Addiction is a Multi-Stage Process: Epidemiological and Behavioural Genetic Analyses of Drug Use Transitions

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Addiction is a Multi-Stage Process: Epidemiological and Behavioural Genetic Analyses of Drug Use Transitions

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'I could tell you my adventures—beginning from this morning,' said Alice, a little timidly: 'but it's no use going back to yesterday, because I was a different person then.'

'Explain all that,' said the Mock Turtle.

'No, no! The adventures first,' said the Gryphon in an impatient tone: 'explanations take such a dreadful time.'

Lewis Carroll, Alice's Adventures in Wonderland and Through the Looking Glass

Abstract

Background Unlike many other psychiatric conditions, individuals with drug dependence pass through a number of intermediate stages before developing a clinical condition. Studying variation in speeds of transitions between these stages provides novel insights into Substance Use Disorder aetiology.

Aims To explore:

1. Relationships between early cannabis transitions and later outcomes;
2. Relationships between individual, childhood, mental health and drug use factors and transitions;
3. The extent to which speed of early cannabis transitions are influenced by genes and environment, and the genetic correlation with dependence;
4. The relationship between early heroin transitions and later heroin outcomes, and the effect of route of administration.

Design Three samples were analysed: 1) 3824 Australian twins and siblings; 2) 93 opiate substitution treatment clients; 3) 408 heroin users.

Methods Cox PH Survival Analysis; Regression Analysis; Classic and Bivariate Twin Modelling.

Findings Early onset of first opportunity to use (OTU) cannabis was associated with increased risks of later cannabis use outcomes, with those who reported first OTU before age 14 being twice as likely to report cannabis daily use, abuse/dependence or treatment-seeking relative to those whose first OTU occurred after age 18.

Faster progression to subsequent cannabis use was associated with increased risks of later cannabis use outcomes, with those whose subsequent use was within a week of initiation being twice as likely to report cannabis daily use, and more likely to abuse/dependence, relative to those whose subsequent use was more than year after initiation.

Conduct disorder, weekly tobacco use, male gender and parental drug problems were associated with faster progression to both OTU cannabis, and from opportunity to dependence. A genetic correlation of .54 was observed between

age of OTU cannabis and cannabis abuse/dependence.
Early OTU heroin was associated with three times likelihood of overdose and neck/groin injecting. Injecting heroin was associated with faster progression to daily use.

Conclusions Considering the stage-sequential nature of drug use has identified factors associated with dependence that also influence behaviour from the earliest stage of drug involvement, and has revealed potential targets for intervention.

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Abbreviations Used in Thesis

Abbreviaiton	Definition
95% CI	**95% C**onfidence **I**ntervals
-2LL	**-2 L**og **L**ikelihood Ratio
A	**A**dditive Genetic Effects
AIC	**A**kaike **I**nformation **C**riterion
ATR	**A**ustralian **T**win **R**egistry
C	Shared (**C**ommon) Environment Effects
CATI	**C**omputer **A**ssisted **T**elephone **I**nterview
CD	**C**onduct **D**isorder
CSA	**C**hildhood **S**exual **A**buse
Cox PH	**Cox P**roportional **H**azards
DALYS	**D**isability **A**djusted **L**ife **Y**ears
df	**D**egrees of **F**reedom
DV	**D**ependent **V**ariable
DZ	**D**i**z**yogtic
E	Unique **E**nvironment Effects
EEA	**E**qual **E**nvironments **A**ssumption
FHAM	**F**amily **H**istory **A**ssessment **M**odule
IAR	**I**nitial Heroin **A**dministration **R**oute
IoPPN	**I**nstitute **o**f **P**sychiatry, **P**sychology and **N**euroscience
IV	**I**ndependent **V**ariable
MDD	**M**ajor **D**epressive **D**isorder
MZ	**M**ono**z**yogtic
OR	**O**dds **R**atio
OST	**O**piate **S**ubstitution **T**reatment
PAI	**P**rivileged **A**ccess **I**nterviewers
QIMR	**Q**ueensland **I**nstitute of **M**edical **R**esearch
s.d.	**S**tandard **D**eviation
SDS	**S**everity of **D**ependence **S**cale
SLAM	**S**outh **L**ondon **a**nd **M**audsley NHS Trust
SNP	**S**ingle **N**ucleotide **P**olymorphism
SUDs	**S**ubstance **U**se **D**isorders
THC	**T**etra**h**ydro**c**annabinol
TVC	**T**ime **V**arying **C**ovariates

CHAPTER 1

Introduction

1.1 What Are Substance Use Disorders?

Drug use is prevalent amongst adolescents; one Australian study has estimated lifetime prevalence of adolescent cannabis use as high as 60% (Patton et al., 2002). It is widely accepted that a large proportion of those using drugs in adolescence will do so infrequently, with a smaller proportion developing Substance Use Disorders (SUDs) and related problems, as defined by DSM-IV and DSM-5 criteria (American Psychiatric Association, 2000; American Psychiatric Association et al., 2013). SUDs require those diagnosed to experience a number of negative effects of drug use, including the neglect of personal relationships, employment and health; significant time investment in obtaining and using the drug; control being lost as the drug is used in greater quantitates, or more frequently, than an individual intends; and unsuccessful attempts to cut down or stop using the drug of abuse. As tolerance towards the drug increases, greater amounts are needed to meet the psychological or physical demands underlying use (American Psychiatric Association, 2000; American Psychiatric Association et al., 2013).

SUDs are an area of global and national health concern (Degenhardt et al., 2014), with prevalence estimated at 24% in the US population (Kessler et al., 2005) and 3.6% in the UK (Fuller et al., 2009). These disorders directly account for 20.0 million Disability Adjusted Life Years (DALYs), equating to 0.8% of global DALYs from all

causes (Degenhardt et al., 2013).

1.1.1 Cannabis and Heroin Use

Prevalence of drug treatment seeking indicates that, of illicit drugs, cannabis and opioids have some of the most negative impacts on the health of those using them (UNODC, 2014). The focus of this thesis is cannabis use and disorders, with some additional consideration of heroin use and outcomes. Cannabis is the most commonly used illicit drug, with prevalence of lifetime use estimated at between 2.7% and 4.9% of the global population aged 15-64 years (UNODC, 2014); although as stated above, prevalence may be much higher amongst adolescent populations (Patton et al., 2002). It is estimated that 10 - 16% of cannabis users develop dependence (Anthony, 2006), and that globally 13.1 million individuals meet criteria for cannabis dependence, contributing 10.3% of the illicit drug use global burden of disease (Degenhardt et al., 2014).

Conversely heroin use is less common, with global prevalence of use of heroin and other opioids estimated at 0.6% - 0.8% (UNODC, 2014). However, despite the low use prevalence, dependence on these drugs accounts for the highest proportion (46%) of illicit drug contribution to DALYs (Degenhardt et al., 2014). Heroin use is also associated with significant health risks beyond addiction, such as overdose, which is the leading cause of mortality amongst users of this drug (Degenhardt et al., 2011).

1.2 The Aetiology of Substance Use Disorders

It could be assumed that the effect of drug pharmacology on brain biology is the main contributor to the development of SUDs. From a behavioural pharmacological viewpoint, the development of dependence is relatively straightforward. The drug creates a “reward” effect in the brain, most likely through activation of the mesolimbic dopamine pathway, and each use of the drug strengthens this association (West and Brown, 2013). After repeated use, physiology can adapt to the drug, resulting in withdrawal when the drug is not used (West and Brown, 2013). If the aetiology of drug dependence were purely pharmacological we could expect all those who initiated use of the drug to follow the same course. However, as

stated above, only 10-16% of those who use cannabis go on to develop dependence (Anthony, 2006). A complete understanding of the aetiology of this disorder requires exploration of the cause of these individual differences.

Genetic variation may contribute to the aetiology of SUDs. Humans share 95% of their genes, with the remaining 5% underlying individual variation. Twin methodologies are one way that the extent to which genetics influence a trait can be examined (Plomin et al., 2013). Agrawal and Lynskey (2008) noted that estimates of the heritability of drug dependence ranged from 0.30 to 0.70, and were broadly equivalent across individual drugs. Genetic influences that increase vulnerability to drug use across drug classes have been identified (Moffitt et al., 2005; Tsuang et al., 1999), although some evidence has been found for separate, albeit highly correlated, licit and illicit drug genetic factors (Kendler et al., 2003).

However, given that not all of the variation in SUDs is attributable to genetic influences (Agrawal and Lynskey, 2008), we can extrapolate that the environment must also contribute to the variance. A highly cited review of risk for adolescent SUDs identified a wide range of contributing factors, including drug availability, economic deprivation, personality factors (such as sensation seeking and impulse control), individual and family attitudes towards drug use, family conflict, childhood antisocial behaviour, academic failure, peer drug use, and early initiation of drug use (Hawkins et al., 1992). Consequently the study of the aetiology of SUDs is complex, requiring understanding of the effect of a number of factors.

1.3 Considering the Multi-Stage Nature of Drug Use

The development of SUDs is a multi-stage process in which a number of specific transitions must occur. In this way, SUDs differ from many other psychiatric conditions. The first stage of drug involvement is having the opportunity to use (regardless of whether the individual uses the drug or not). Opportunity is required for use to occur, and forms an individual's earliest necessary condition from which they are at risk of developing dependence (Wagner and Anthony, 2002). Following this is initiation of use, progression to subsequent use, and using the drug regularly. Regular use itself can be defined as use within a time period, for example monthly, weekly, or daily. By necessity, only individuals who pass through these stages can progress to the development of SUDs. After the development of SUDs, further

transitions may take place, such as those into treatment and recovery, and in some instances back to drug use. An outline of the stages of drug use is provided in Figure 1.1.

Figure 1.1: The Stages of Drug Use

As stated above, not all individuals who initiate drug use – or who reach some of the subsequent stages of drug use - progress to SUDs. Despite this, studies of dependence commonly compare individuals who are drug dependent against those who are not dependent. This fails to distinguish between non-cases who may have never used a drug and those who used the drug (potentially regularly) but did not progress to disorder development. This conflation of stages leads to uncertainty as to the points in the development of drug dependence at which specific genetic or environmental influences are most prominent (Nelson et al., 2013).

Research that has considered the multi-stage nature of drug use has suggested that the relative strength of genetic and environmental influences varies by both stage of drug use and by developmental age (Distel et al., 2011; Heath et al., 1999; Kendler et al., 1999b; Lynskey et al., 2012; Sartor et al., 2007, 2008). For example, there is evidence from studies of tobacco and alcohol use that suggest that, in their aggregate, genetic influences are relatively weaker at earlier stages in the development of drug dependence (e.g. initiation of use; see Lynskey et al. (2010) for review). Although weak at these early stages, there is evidence that the genetic factors that influence initiation are at least partially shared with those that influence later problem use (Kendler et al., 1999a; Neale et al., 2006b; Heath et al., 2002).

The extent to which the importance of measured genetic and environmental influences on drug dependence may vary across stages of drug involvement deserves further exploration. Early stages of drug use, such as initiation, may be genetically influenced through personality traits such as novelty seeking (Laucht et al., 2007). Drawing on existing research, we can speculate that at subsequent stages, such as drug dependence and development of withdrawal, genetic influences on drug metabolism, or from genes that are associated with use of specific drugs may be more influential (Dick et al., 2014). Similarly, there may be distal (influences which are removed from the onset of behaviour)and

some proximal (influences that may act directly on the onset of that behaviour) environmental risk factors that are unique to specific stages of drug involvement, while others may act across multiple stages, or show correlation while not being identical (Lynskey et al., 2010). Consequently, it is expected that utilising the multi-stage approach will reveal that observed genetic and environmental associations alter throughout the development of SUD.

Some recent research utilising the multi-stage approach to drug use has demonstrated differences in association by stage of use. Sartor et al. (2007) reported a number of factors that were unique to onset of alcohol use (e.g. male gender, attention deficit hyperactivity disorder, parental divorce and maternal alcohol dependence), while others were unique to the transition from alcohol use to dependence (e.g. nicotine dependence, cannabis abuse, generalised anxiety disorder). Differences have also been observed for genetic risk factors by stage of drug use. Belsky et al. (2013) reported that a multi-locus genetic risk score, derived from the results of meta-analyses for nicotine dependence, was unrelated to initiation of tobacco use but was significantly associated with increased risks for daily tobacco use, more rapid progression from initiation to heavy use, increased risks for the development of nicotine dependence and reduced likelihood of successful cessation.

There is some evidence that the effect of genes alters across drug use behaviour trajectories. This has been observed in relation to *GABRA2* and *OPRM1*, respectively a gene associated with the inhibitory neurotransmitter GABA, and a gene that encodes opioid receptors. Modelling development and changes in alcohol use over time has found that *GABRA2* is associated with an increase in drunkenness between ages 18–19, suggested to be due to the enhanced independence related to reaching adulthood (Dick et al., 2014). Similarly, *OPRM1* has been found to differentiate those who were light drinkers from those who had progressed to moderate drinking in participants followed up over 6 years (participants on average aged 14.3 at start of study) (van der Zwaluw et al., 2014).

In summary, studies utilising the multi-stage approach are likely to reveal that differential influences, both internal and external, operate at each stage. However, despite the utility of taking a multi-stage approach, this is rarely incorporated into studies of drug use. In this thesis I apply the multi-stage approach to gain new insights into the aetiology of SUDs.

1.4 Exploring SUD Aetiology Through Studying the Speed of Drug Use Transitions

Variation in drug use progression can be used to explore the relationship between the earlier stages of drug use and SUDs. Speed of transition is one point of variation that can be utilised for this purpose. The majority of research utilising speed of transitions in drug use comes from the literature on early initiation of use (with younger age at initiation of use representing a faster transition to use). By taking a selection of studies from this research area, focussed on initiation of cannabis or heroin use, I will provide an overview of what current knowledge of speed of early drug use transitions reveals about SUD aetiology.

A number of studies of early initiation of use have identified a relationship to later drug use outcomes. In a cross-sectional US study of 42,862 participants age 18 and over, the prevalence of lifetime drug (including cannabis) abuse and dependence declined as a function of increasing age at onset of drug(including cannabis) use (Grant and Dawson, 1998). Amongst 1520 individuals involved in a longitudinal Australian cohort study, adolescents who experienced early initiation of cannabis use (before age 16) had increased likelihood of experiencing cannabis dependence by age 24 (OR 2.7, 95% CI 1.5-4.8), compared to those who reported later initiation of use (Swift et al., 2008). In the same study, there was evidence of an effect of use frequency. Amongst those in the sample who had initiated cannabis use by age 15, the likelihood of cannabis dependence by age 24 increased with frequency of use at age 15. Similarly, in a birth cohort of 1265 New Zealand children followed-up until 18 years of age, the prevalence of cannabis abuse/dependence at age 16-18 was twice as high amongst those who had used cannabis 10+ times at age 15-16, compared to those who had used cannabis fewer than 10 times (Fergusson and Horwood, 1997). Amongst heroin users, younger age of heroin initiation has been found to be associated with likelihood of overdose (Lynskey and Hall, 1998). These findings indicate a consistent association between earlier age of initiation and later abuse, dependence and other drug use outcomes.

Twin studies have provided evidence that genetic factors influence the age of cannabis initiation, and have provided further insight into the relationship between this and later SUDs. A longitudinal study of 231 African-American female twins tested the genetic contribution to the age of cannabis initiation, and found that

the majority of the variance in age of initiation (with early initiation defined as $\leq$16 years old) was attributable to genetic effects (0.77, 95% CI: 0.77–0.79) (Sartor et al., 2009a).

Genetic effects have also been considered in discordant twin studies, which (due to the genetic and environmental similarity of the twin pairs) allow for genetic effects to be controlled for. In a sample of Dutch twins discordant for early onset of use ($\leq$18 years), likelihood of lifetime regular cannabis use was *not* increased in the early-onset twin compared to the later onset twin (Lynskey et al., 2006). A similar study comparing 311 co-twins discordant for early initiation of cannabis use from an Australian sample, whereby only one twin in each pair initiated cannabis use $\leq$17, found that the twin who reported early initiation of cannabis use was at increased likelihood of later cannabis dependence (OR 1.96, 95 CI 1.25-3.09) (Lynskey et al., 2003); but this association disappeared when analyses were restricted to twin pairs who had both initiated cannabis use. Given that the association to problematic use did not remain after controlling for genetic and environmental similarity, these findings suggest that the association between early onset and later problem use is due to shared genetic and environmental factors.

A large number of individual and external factors that are associated with early initiation of cannabis use have been identified in the literature. The studies of adolescents discussed here have been selected due to their sample size (1000+ participants) and prospective design. Use of cannabis by age 14-15 was associated with having divorced/separated parents (OR 1.5, 95% CI 1.1 – 2.1), peer cannabis use (OR 12.0, 95% CI 8.6–17), daily smoking (OR 4.7, 95% CI 3.5–6.4), high dose drinking (OR 3.2, 95% CI 2.3–4.3), and anti-social behaviours (OR 3.9, 95% CI 2.8–5.5) (Coffey et al., 2000). Similarly, initiating cannabis use before age 14 was significantly (P=$\leq$0.001) correlated with parental report of disruptive behaviour (r=0.14), oppositional problems (r=0.11), attention deficit hyperactivity (r=0.12) and conduct problems (r=0.14) (Creemers et al., 2009a). High childhood hyperactivity-inattention symptoms and high Conduct Disorder (CD) symptoms have been associated with speed of transition to initiation of cannabis use (males HR 1.95, 95% CI 1.04-3.64, females HR 2.01, 95% CI 1.31–3.09) (Galéra et al., 2010). Sensation seeking personality traits were associated with early cannabis use, represented as use before age 13 (OR 1.22, 95% CI 1.05–1.43) (Creemers et al., 2009b).

The literature on early initiation of cannabis and heroin use, a selection of which

is considered above, highlights the range of factors that contribute to variation in speed of transition and the potential for speed of transition to be indicative of later drug use outcomes. However, there are issues with using this literature to explore transition speed. Firstly, the majority of studies do not use a continuous measure of time to initiation, resulting in wide heterogeneity in what is considered "early" initiation (and, consequently, what is considered a fast transition). Within the studies cited above, the cut-off point for early initiation ranges from 13 – 17 years of age. A large number of developmental changes can be expected to occur between these time points, which limits the extent to which results can be generalised. Consequently, in order to isolate speed of transition from individual age, the study of transitions needs to focus on speed between stages. Opportunity to use a drug has been recognised as the first stage of drug use, and consequently studies of age of initiation (rather than time form opportunity to initiation) do not strictly represent between-stage transitions. Secondly, there is wide variation in the factors that have been considered in relation to the speed of transition. This makes it difficult to compare factors influencing early initiation with factors influencing the development of SUDs, which may give an indication of what underlies the association between early initiation and later drug use outcomes.

Therefore, whilst the research focussed on age of initiation of use can give some insight into SUD aetiology, more meaningful and informative results will be gained by considering literature on other multi-stage transitions, and studies that report on transitions between stages. Considering multiple stages will allow identification of how influences of specific risk factors may change between stages, providing a clearer picture of what may influence progression towards dependence.

The focus placed on cannabis and heroin in this thesis adds an additional dimension to the issue of speed of transition. The majority of research examining transitions in drug use has focussed on alcohol (Donovan and Molina, 2011; Wu et al., 2006; Pitkänen et al., 2005; Johnson et al., 2000; Hingson and Zha, 2009; Hingson et al., 2006, 2008; Dube et al., 2006; Grant et al., 2006; Schulenberg and Maggs, 2002) and tobacco (Maes et al., 2004a; Stallings et al., 1999; Jamal et al., 2011; Kendler et al., 1999b); both licit and widely-available drugs in most countries. It is of interest to observe whether similar patterns emerge for transitions in illicit drug use. Cannabis and heroin are of interest as they are understudied in the field, and in addition differ from each other in terms of prevalence of use (see

Section 1.1.1, above). Additionally, those using heroin have a high proportion of reported childhood abuse and neglect compared to populations using other drugs (Darke, 2011). Consequently focussing on these drugs provides a test of whether associations with transition speed apply to illicit drugs, to drugs with differing prevalence of use, and to populations with different degrees of dysfunction.

1.5 Literature Review: Outcomes and Factors Associated with the Speed of Specific Transitions in Cannabis and Heroin Use

1.5.1 Search Strategy

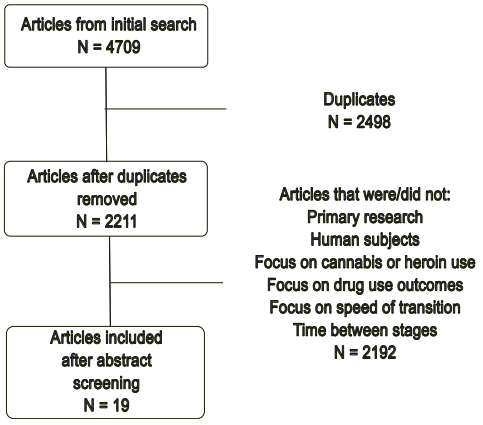
Table 1.1: Literature Review Search Terms

Drug	Stages	Transition Speed
Cannabis	Opportunity	Fast
Marijuana	Initiation	Slow
Hashish	First use	Early
Opiate	Onset	Rapid
Heroin	Regular use	Late*
	Subsequent use	Speed
	Second use	
	Daily use	
	Weekly use	
	Monthly use	
	Stage	
	Transition	

A systematic search was conducted to gain a comprehensive view of the existing research exploring the specific transitions represented in a multi-stage model of drug use progression (see Figure 1.1), in populations of cannabis or heroin users. The focus of the review was on factors influencing transition speed, and the relationship to drug use outcomes. Any research on speed of transition between stages of drug use (or age at opportunity, given that there is no preceding stage of drug involvement) was included. Search terms (abstract and keyword search) were combined to identify the drugs of interest, stages of drug use, and transition speed (see Table 1.1 for list of terms).

Only primary journal articles written in English and using human participants were screened. See Figure 1.2 for the search numbers and screening criteria. A total of 19 articles were identified that focussed on speed of transition between the stages of drug use. See Table 1.2 for search results grouped by drug and transition, and a quality rating for each paper (see Appendix 1 for Quality Rating Table).

Figure 1.2: Search Results and Exclusion Criteria



The transitions that emerged from the literature review are presented in Figure 1.3. The majority of research focuses on the transition from initiation to problematic use, or abuse/dependence; these were studied in 9 papers. As Figure 1.3 demonstrates, transitions from initiation to problem use encompass a number of other stages of drug use. Opportunity (represented as age of onset, given that there are no preceding stages of drug use) was reported on in 8 papers, and progression from initiation to regular use was studied in 5 papers. The majority of the research was taken from literature focused on cannabis use, which was featured in 14 identified papers; only 7 papers explored transitions in heroin use. This likely reflects the low prevalence of heroin use, and the relatively high prevalence of cannabis use, in general populations. The findings of the review are summarised below.

1.5.2 Critical Appraisal of Identified Studies

Five papers that were identified in the literature had quality score lower than 10. For three of these papers (Reboussin et al., 2007; Best et al., 2005; Chamla et al., 2006), the focus of the research was not on transition speed. Consequently findings drawn

from these papers may be limited, and the approach to analysis of the variables is unlikely to have been given careful consideration. In keeping with this, one of these papers (Reboussin et al., 2007) applied an approach whereby opportunity by 8th grade was compared against no opportunity by this stage. This does not allow differentiation between those who had later opportunity and those who never had opportunity, which may conflate estimates within the analysis.

Three papers had a sample size that under 500 participants or a response rate of fewer than 60% (Reboussin et al., 2007; Haas and Peters, 2000; Ridenour et al., 2006; Chamla et al., 2006). These factors may lead to a sample that is unrepresentative of the population of interest, and this should be considered when attempting to extrapolate their findings to the wider population. Similar consideration is required for the studies that utilised convenience sampling rather than random sampling of the population of interest (Best et al., 2005; Ridenour et al., 2006).

Two of the low quality studies utilised analyses methods that did not allow for the adjustment of confounding variables (Haas and Peters, 2000; Ridenour et al., 2006). Consequently the findings presented from these studies may not provide accurate estimates of observed associations. For the low quality studies that did not make use of prospective data (Haas and Peters, 2000; Chamla et al., 2006), there is potential for recall bias to affect observations.

Table 1.2: Quality ratings of literature on speed of transitions in heroin and cannabis use, identified through systematic search

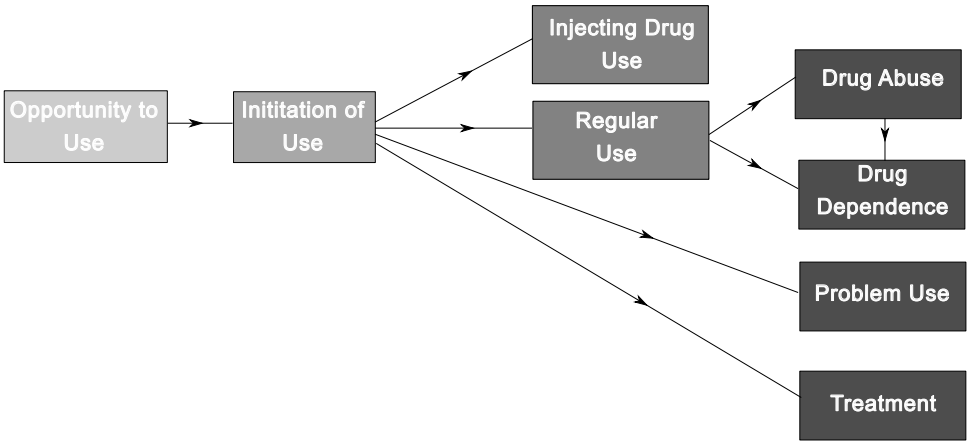
Transition	Drug	Reference	Study design	Selection method	Study focus	Prospective	Control group	Measure of transition speed	Analysis	Sample Size	Response rate >60%	Quality score (max = 16)
Age of Opportunity	Cannabis	Reboussin et al 2007	2	2	0	1	1	0	1	0	1	8
		Agrawal et al 2013	2	2	1	0	2	1	2	2	1	13
		Best et al 2005	2	0	0	0	2	1	1	2	1	9
		Chen et al 2005	2	2	0	1	2	1	2	2	1	13
		Storr et al 2011	2	2	1	1	2	1	2	2	1	14
		Van Etten & Anthony 1999	2	2	0	0	2	2	0	3	1	12
		Lynskey et al 2012	2	2	0	0	2	1	2	2	0	11
	Heroin	Van Etten & Anthony 1999	2	2	0	0	2	2	0	3	1	12
Opportunity to Initiation	Cannabis	van Etten et al 1999	2	2	0	0	2	2	0	3	1	12
		van Etten et al 1997	2	2	0	0	2	2	0	3	1	12
		Van Etten & Anthony 1999	2	2	0	0	2	2	0	3	1	12
	Heroin	van Etten et al 1999	2	2	0	0	2	2	0	3	1	12
		Van Etten & Anthony 1999	2	2	0	0	2	2	0	3	1	12

Table continued on next page

Transition	Drug	Reference	Study design	Selection method	Study focus	Prospective	Control group	Measure of transition speed	Analysis	Sample Size	Response rate >60%	Quality score (max = 16)
Initiation to Regular Use	Cannabis	Ridenour et al 2006	2	0	1	1	2	2	0	1	0	9
	Heroin	Mills et al 2004	2	1	1	0	2	2	1	1	1	11
		O'Keefe et al 2016	2	0	1	1	2	2	2	1	1	12
		Stoltman et al 2015	2	0	1	0	2	2	0	2	1	10
		Woodcock et al 2015	2	0	1	0	2	2	0	2	1	10
Initiation to Problem Use	Cannabis	Haas & Peters 2000	2	1	1	0	2	2	0	0	0	8
		Ridenour et al 2006	2	0	1	1	2	2	0	1	0	9
		Sartor et al 2015	2	2	1	1	2	2	2	2	1	15
		Sartor et al 2013a	2	2	1	1	2	2	2	2	1	15
Initiation to Abuse/ Dependence	Cannabis	Behrendt et al 2009	2	2	1	1	2	2	2	2	1	15
		Chen et al 2005	2	2	0	0	1	2	1	3	1	12
		Ridenour et al 2006	2	0	1	1	2	2	0	1	0	9
	Heroin	Ridenour et al 2005	2	2	1	0	2	2	0	2	1	12
Abuse to Dependence	Cannabis	Ridenour et al 2005	2	2	1	0	2	2	0	2	1	12
Initiation to Injecting	Heroin	Chamla et al 2006	2	1	0	0	2	2	2	0	0	9
Initiation to Treatment	Heroin	Mills et al 2004	2	1	1	0	2	2	1	1	1	11

For paper quality rating criteria, see Appendix 1)

Figure 1.3: Transitions Through the Stages of Cannabis/Heroin Use Studied in the Speed of Transition Literature



1.5.3 Age of Opportunity

Cannabis Research

Opportunity to use is the stage transition with the most research, and the only stage transition that has been considered in relation to later drug use outcomes. In a sample of 2078 14-16 year old school pupils, age at first opportunity to use cannabis was not significantly associated with frequency of past-month cannabis use (Best et al., 2005). However, there was a significant negative correlation between age of first opportunity to use cannabis and frequency of use ($r=0.32$, $P \leq 0.001$), before regression adjustment for age at tobacco and alcohol opportunity and initiation, and age at cannabis initiation. This association may have been over adjusted; the stated covariates have potential for collinearity with age of opportunity, and it is plausible that age of initiation may mediate rather than confound this association. Exploring the relationship between opportunity and later outcomes was not a main aim of this study. Analyses focused specifically on age of opportunity to use and the associations with drug use outcomes, taking a careful approach to confounding variables, may yield different results.

Onset of this stage tends to happen in adolescence, and consequently identified associated factors focus on early life influences. Results from a longitudinal study following individuals from first grade of school to early adulthood (N=1692) identified that low parental involvement (RR 1.4, 95% CI 1.1–1.7), but not low levels

of parental monitoring (RR 1.0, 95% CI 0.8–1.2) or coercive discipline (RR 1.2, 95% CI 1.0–1.5), was associated with having the opportunity to use cannabis before age 19-20 (Chen et al., 2005b). Having opportunity to use cannabis by age 14 has been found to be associated with aggressive/disruptive behaviour, deviant peer affiliation, and childhood disadvantage (Reboussin et al., 2007), when compared to individuals who had not had the opportunity to use cannabis by this age. A study looking at similar factors, taking behavioural measures when participants were a mean age of 6.5 years old and measuring cannabis opportunity at a mean age of 21, found gender differences in the factors influencing earlier opportunity to use cannabis (Storr et al., 2011). Earlier opportunity to use cannabis was associated with higher childhood aggressive/disruptive behaviour in males but not females (rank P=0.02; mean age 14.2 for males with higher levels of aggressive/disruptive behaviour mean age of 14.7 for males with lower levels), higher reading scores in females ($P \leq 0.01$; mean age 14.7 for those in quartile 3, mean age of 15.6 for females in the lowest reading readiness quartile), and higher maths readiness in males (mean age 14.7 for males in quartile 1, mean age of 14.1 for those in quartiles 3 or 4; P=0.01 and 0.02 respectively). It is important to note that no further behavioural or contextual measures were assessed between ages 6.5 and 21. This is a long time period in which a number of developmental and situational changes will occur for individuals, and the result is that a number of potential influencing variables were unable to be considered.

Gender differences in age of opportunity to use cannabis have not been identified. Research from a large (N = 131226) representative cross-sectional survey in the USA found mean age of first opportunity to use cannabis was the same for males and females (16 years of age for both males and females, P=0.86) (van Etten and Anthony, 1999). With regard to other factors, the use of tobacco has been studied in relation to age of opportunity to use cannabis. In a large, cross-sectional Australian twin sample (N=3797), those who had lifetime history of regular tobacco use (smoking 100+ cigarettes) were found to have an earlier opportunity to use cannabis (Agrawal et al., 2013). However, as the study considered *lifetime* tobacco use, rather than tobacco use prior to opportunity to use cannabis, it is not possible to assess the direction of the relationship between regular tobacco use and earlier cannabis use opportunity.

Genetic influences have been observed on age of opportunity to use cannabis.

In one Australian twin study, having early opportunity to use cannabis ($\leq$15 years) had a heritability estimate of 0.72 (CI 0.63–0.80) and unique environment estimate of 0.28 (CI 0.20–0.38) (Lynskey et al., 2012). This indicates that variation in the speed of transition to cannabis use opportunity is predominately attributable to additive genetic factors.

Heroin Research

The only research regarding heroin opportunity focuses on gender differences, which were not identified. Research from a large (N = 131226) representative cross-sectional survey in the USA found that mean age differed for heroin opportunity, but this difference was not statistically significant (18 years of age for males and 17 years of age for females, P=0.11) (van Etten and Anthony, 1999).

1.5.4 Opportunity to Initiation

Cannabis Research

Very little research has focussed on the transition from opportunity to initiation of drug use. The review identified three papers that had explored this transition, based on the same study that utilised data from a large representative cross-sectional survey in the USA conducted every three years from 1979 - 1994 (total N = 131226). A rapid transition from opportunity to initiation of use was observed in 66% of those who initiated cannabis use (van Etten and Anthony, 1999).

One paper, again based on the same sample, reported basic proportions and identified that those who had younger age of opportunity to use cannabis had a slower transition to initiation of use. Of participants who reported opportunity to use cannabis at age 12, 31% reported initiating cannabis use within a year, whereas this proportion rose to 52% for those reporting opportunity at age 21 (van Etten et al., 1997). Further statistical tests had not been conducted on these figures so significance of this difference cannot be determined.

Cohort differences were not identified in speed of this transition. The proportion progressing to initiation within a year of their first opportunity increased slightly between 1982 – 1994 for cannabis use, but remained relatively stable (van Etten and Anthony, 1999; van Etten et al., 1997). Again, further statistical tests were not

conducted on these figures so significance cannot be determined. No significant gender differences in the proportion of participants making a “rapid” transition (within the same year) from opportunity to initiation of cannabis use at any survey year (van Etten et al., 1999).

Heroin Research

A study that utilised data from a large representative cross-sectional survey in the USA conducted every three years between 1979 - 1994 (total N = 131226) identified a rapid transition from opportunity to initiation of use was observed in 84% of those who initiated heroin use (van Etten and Anthony, 1999). Consequently, it is likely that defining transition from opportunity to initiation within a year as a “rapid” transition is not accurate. Rather, it reflects the majority of individual transitions. Exploring this transition in terms of weeks/months may be more appropriate.

Cohort differences were not identified in speed of this transition. The proportion progressing to initiation within a year of their first opportunity increased slightly between 1979 – 1994 for heroin use, but remained relatively stable (van Etten and Anthony, 1999; van Etten et al., 1997). Gender differences in transitions speed were observed for heroin in 1979 and 1994 (a higher proportion of males making a fast transition) (van Etten et al., 1999).

1.5.5 Initiation to Regular Use

Cannabis Research

A study of 590 participants, recruited in childhood to represent groups at different levels of risk for substance involvement, found no significant differences between males and females in speed of transition from initiation of cannabis to monthly cannabis use (Ridenour et al., 2006).

Heroin Research

The majority of identified heroin research is focused on the transition from initiation to “regular” use. Two of these studies have focussed on age as a factor in the speed of this transition. One study of 615 Australian heroin users found that those aged 18-24 at time of interview had experienced a more rapid transition from heroin initiation

to using at least once a month compared to those aged over 24 in the sample (0.4 vs. 1.1 years, P≤0.001) (Mills et al., 2004). The study authors suggest this may reflect an increase in drug availability, and changes in societal attitudes towards drug use. However, given that this is a population of "young" heroin users it is possible that this group reflects a distinct population amongst those in treatment. Considering the characteristics of the complete sample, the mean age at first seeking treatment was 23 (Ross et al., 2003). The majority (89%) of the 18-24 heroin users were in treatment, and whilst information on the proportion seeking treatment before age 23 is not provided it is likely that many of this 18-24 sample sought treatment earlier than the base population mean. Additionally, compared to the base population the 18-24 year olds had a lower mean age of heroin initiation (18-24 = 16.8, s.d. 2.4, whole sample =19.7, s.d. 5.3) and initiation of injecting (18-24 = 17.8 s.d. 2.7, whole sample = 20.3, s.d. 5.4). It is possible that these differences, or factors relating to them, have also contributed to the more rapid transition to monthly use amongst 18-24 year olds in the sample. Supporting these findings, a separate study of 562 out-of-treatment USA heroin users found that younger age at initiation of heroin use was associated with more rapid progression from initiation to using heroin at least three times per week (Woodcock et al., 2015).

The review has also identified that the very use of heroin, as opposed to other drugs, may lead to comparatively faster transition to regular use. Amongst a sample of 691 injecting drug users, recruited through street outreach and snowball sampling in Melbourne, those injecting heroin had a more rapid transition from first injecting heroin to injecting once a month (1.28, 95% CI 1.09–1.50) compared to those injecting any other drugs (O'Keefe et al., 2016). The study authors identify that heroin is associated with greater severity of dependence compared to methamphetamine (the other predominantly used drug in the sample), and these results indicate that speed of transitions in drug use can be influenced by drug pharmacology.

Finally, a demographic effect on speed of this transition has been identified. A study of 554 out-of-treatment USA heroin users found that Caucasian Americans had a faster progression than African Americans from initiation of heroin use to using more than three times per week (interaction significant at P≤0.01 for both males and females) (Stoltman et al., 2015). This study did not find a significant effect of gender on speed of progression to regular heroin use.

1.5.6 Initiation to Injecting

Heroin Research

One study has considered the heroin-specific transition of progression from non-injecting to injecting route of administration (ROA) using a sample of 266 Chinese heroin-using participants recruited from detoxification treatment. The majority of participants had begun using heroin through nasal ROA, and time from initiation of use to injecting heroin was associated with age (HR 0.91, 95% CI 0.89–0.93) and duration of heroin use (HR 1.14, 95% CI 1.07–1.21) (Chamla et al., 2006). This may reflect cohort effects on speed of transition.

1.5.7 Initiation to Problem Use

Cannabis Research

The research on the transition from initiation to problem drug use provides further evidence of genetic effects on transition speed. In a sample of 4193 female participants (comprised of a mix of twins and sibling pairs), a sibling experiencing cannabis use disorder symptoms was associated with increased hazard of progression to cannabis use disorder at any age in African-American participants (HR 1.91, CI 1.34-2.72). Analyses were stratified by onset age for European Americans, and sibling experience of cannabis use disorder symptoms was associated with participant cannabis use disorder symptom onset at age 18-21 (HR 2.50, CI 1.81-3.45) and at age 21+ (HR 2.23, CI 1.27-3.90) (Sartor et al., 2015). Although the study design does not allow these findings to be disentangled from the environmental influences shared by siblings, they hint at a shared genetic influence between cannabis use disorder symptoms and increased rate of progression between initiation and problematic use.

A number of proximal and distal factors have been found to be associated with the speed of this transition. Childhood Sexual Abuse (CSA) has been associated with rate of progression from cannabis use initiation to problematic use. In a sample of 4193 twin and sibling pairs, CSA was associated with increased hazard of progression to cannabis use disorder after 21 years of age in African American participants (HR 2.17, CI 1.31-3.59), and at any age in European-American participants (HR 1.51, CI 1.21-1.88) (Sartor et al., 2015). In a sample of twins

and family members at risk of drug use (N=4102) the rate of progression to first symptom of cannabis use disorder was increased amongst those who experienced CSA (HR 1.40, CI 1.11–1.77) (Sartor et al., 2013a).

Speed of transition to problematic cannabis use has also been associated with mental health factors. Using a sample of 4102 female twin and sibling pairs, rate of progression from initiation of cannabis use to problem use was increased amongst those who experienced Conduct Disorder (CD) (HR 1.82, CI 1.41–2.35) and Major Depressive Disorder (MDD) (HR 1.30, CI 1.03–1.65) (Sartor et al., 2013a). Due to the methodology used, the measure of MDD was only included in the analysis if onset was reported before the development of problematic use, indicating that this factor may have a causal relationship with speed of transition.

In the same sample, those who were regularly smoking tobacco at least once a week for 2 months prior to problematic cannabis use (and had smoked greater than 20 cigarettes in their lifetime) were found to have faster progression from initiation to their first symptom of problematic use (HR 2.02, 95% CI 1.58 - 2.64) (Sartor et al., 2013a). Those who had lifetime alcohol use had increased rate of progression to their first symptom of problematic cannabis use (HR 1.95, CI 1.27–2.99).

The research on this transition has not identified any effect of gender on the speed of progression. No significant differences were observed between males and females for latency from initiation to problematic marijuana use in a sample of 160 poly-substance using individuals (Haas and Peters, 2000). Similarly, no significant differences between males and females were observed in speed of transition from cannabis initiation to first problem in a sample of 590 participants recruited in childhood to represent groups at different levels of risk for substance involvement (Ridenour et al., 2006).

1.5.8 Initiation to Abuse/Dependence

Cannabis Research

Analysis of this transition has focussed on the factors that are associated with faster progression to abuse or dependence. No significant differences between males and females have been observed in speed of transition to dependence (Ridenour et al., 2006). Amongst 3352 participants who reported cannabis use for the first time within the 24 months prior to assessment, drawn from a

large US survey on drug abuse (residents aged 12 and over, total N = 114241), those who reported using three or more drugs (from a choice of tobacco, alcohol, cocaine/crack, heroin, hallucinogens, inhalants, pain relievers, anxiolytic tranquilizers, stimulants other than cocaine, and sedative-hypnotics) were found to have a fast progression from cannabis initiation to dependence (RR 2.2, 95% CI 1.1–4.3) (Chen et al., 2005a). In this study, a progression was considered fast if the individual had developed dependence within 24 months of initiation. In the same study, age of first cannabis use was associated with fast onset of dependence for the following initiation age groups:16–17 (RR 9.4, CI 2.2–40.0); 14–15 (RR 13.2, CI 3.2–55.5); 11–13 (RR 11.6, CI 2.7–49.9). This is in contrast to a longitudinal study of German youths (N = 3021), which found that those with later onset cannabis use had faster progression to cannabis abuse, but slower progression to cannabis dependence (Behrendt et al., 2009). However, the approach to analysis may have affected these German results. By stratifying the analysis by age of initiation, some younger age groups had lower power (N = 130). Applying age of onset data as a continuous, non-stratified variable may have provided a more comprehensive picture of the progression to abuse and dependence.

1.5.9 Abuse to Dependence

Cannabis Research

One study has considered the speed of transition from the development of drug abuse to dependence. Using a sample of 1226 participants recruited from clinical and community groups for the DSM-IV field trials, the study identified no significant differences in length of time from meeting cannabis abuse criteria to meeting cannabis dependence criteria between early cannabis initiators (the 25% of the study population who reported youngest ages of cannabis use) and later initiators, between men and women, or between African-Americans and Caucasian Americans (Ridenour et al., 2005). However, there are issues surrounding the study of this transition. In this study, it was observed that a proportion of the sample (16% of males and 28% of females) met criteria for cannabis dependence before meeting criteria for abuse; as such, there could not be a length of progression from abuse to dependence. Abuse and dependence have been found to have a single underlying continuum of severity (Lynskey and Agrawal, 2007), and this is reflected

in the DSM-5 (released since the publication of the Ridenour et al. (2005) study) whereby drug abuse and dependence have been combined into a measure of SUD (American Psychiatric Association et al., 2013). The "flipped" order of progression seen in the described study highlights that the onset of these symptoms will not necessarily be sequential, and demonstrates the importance of careful, sequential application of the multi-stage approach to drug use.

1.5.10 Initiation to Treatment

Heroin Research

The only research relating to time to treatment seeking was conducted in a heroin-using population. One study of 615 Australian heroin users found that those aged 18-24 at time of interview had experienced a more rapid transition from heroin initiation to treatment for their heroin use (2.4 vs. 5.1 years for those aged 24+ at interview, $P \leq 0.001$) (Mills et al., 2004). As discussed above, compared to the base population the 18-24 year olds had a lower mean age of heroin initiation (18-24 = 16.8, s.d. 2.4, whole sample =19.7, s.d. 5.3) and injecting (18-24 = 17.8 s.d. 2.7, whole sample = 20.3, s.d. 5.4), which could have contributed to the more rapid transition to treatment seeking in this sample. Additionally, being female was independently associated with an increased speed of progression from initial heroin use to first treatment for heroin use (HR= 1.35, 95%CI 0.57–2.13).

1.5.11 Literature Review Conclusions

A number of proximal and distal factors have emerged from the literature as associated with speed of transition through one or more of the stages of drug use. For cannabis transitions these include childhood disruptive behaviour, strict parenting, CSA, MDD, CD, and pre-existing tobacco or other drug use. One factor that emerged as consistently *not* associated with transitions in cannabis use was gender. Fewer factors have been explored in relation to heroin, but for this drug gender and ethnicity were found to have some influence. Additionally for heroin there were indications of cohort effects on use patterns, although the underlying reasons for these were not explored.

Some evidence for genetic effects on speed of transition emerged from this review. A high heritability estimate was observed for age of opportunity to use cannabis (Lynskey et al., 2012), and other genetically informative designs indicate there are genetic effects on speed of progression to problematic use. Heritability estimates for speed of transition are high even at the early stages of drug use, given that studies of the stages of drug use (but not focussed on the speed of transitions) typically identify lower heritability estimates at earlier stages (Lynskey et al., 2010).

Finally, the review has highlighted that there may be transitions specific to use of individual drugs. Research into heroin use has considered transitions in ROA, which is significant for this drug given the increased health risks associated with moving from non-injecting ROA to injecting (Strang et al., 1998). This demonstrates the importance of considering the idiosyncrasies of different drugs when exploring transitions.

1.5.12 Identified Gaps in the Literature

One of the most striking findings is the dearth of literature focussing on the relationship between speed of transition and later drug use outcomes in either cannabis or heroin use. Only one identified study explored this topic (Best et al., 2005). This is especially surprising given the research linking early initiation of cannabis use with later problem use outcomes. The time between drug use initiation and the development of SUDs has been found to be short, leaving only a small window for targeted intervention (Wittchen et al., 2008). Therefore, exploring whether early transitions in drug use are related to later problems is an avenue that has scope for improving prevention methods.

The majority of the transitions that were identified were those from initiation of drug use to varying degrees of problematic use. Given the importance of identifying early markers of SUDs (Wittchen et al., 2008), there is utility in exploring more transitions from the early stages of drug use. Considering drug use from a multi-stage approach identifies that there are transitions that have not yet been explored. One is the transition from initiation to subsequent use (see Figure 1.1). Exploring a novel transition such as this will further break down the progression towards drug dependence, and has the potential to develop new understanding of the processes underlying the development of dependence.

Existing findings indicate that individual, childhood, and mental health factors play a role in speed of transition. However, currently the factors studied have only been sparsely investigated across the different stages of drug use. Testing the association between a consistent selection of factors and different stage transitions would provide greater understanding of how influences alter as an individual progresses towards drug dependence.

Research has indicated that genetic factors play a role in speed of transition, but as this has only been explored in relation to a small number of transitions a large amount still remains unknown about the genetic contribution to variation in speed. It is currently unknown whether genetic influences on transition speed have a consistent contribution, or if this differs across transitions. Additionally, research has failed to explore whether the genetic factors contributing to speed of transitions in drug use overlap with those contributing to the development of SUDs. Further exploration of this area may have implications for our understanding of the aetiology of drug dependence.

The literature search focussed on cannabis and heroin use, but very little research was found relating to heroin use transitions. Additionally, the research identified that users of heroin may experience faster transitions in drug use than those using other drugs. Consequently it is possible that associations between transition speed and heroin use will differ from those seen in cannabis using populations. There is scope to explore the association between transition speeds and outcomes in heroin use, and to take into account the idiosyncrasies of this drug, such as the importance of ROA and the risk of overdose, when doing so.

1.6 Key Methodologies for Studying Transitions Through the Stages of Drug Use

Studying the genetic, proximal and distal factors that influence speed of transitions in drug use requires the use of methodologies that are 1) appropriate for analysing temporal (time-to-event) data and 2) are able to account for the multi-stage nature of drug use, whereby progression to a stage is contingent on an individual having reached an earlier stage.

1.6.1 Epidemiological Methods

For the study of factors influencing speed of transition, Cox's Proportional Hazards Survival Analysis is an appropriate methodology. Using time-to-event data to explore the effect of factors on an individual's outcome, it has previously been applied in the drug and alcohol research field to study the speed with which an individual develops an outcome such as opportunity to use, initiation of use and development of use disorders (Agrawal et al., 2006; Behrendt et al., 2012; Sartor et al., 2013b; Storr et al., 2011; Waldron et al., 2014), and was used in 9 of the papers identified in the literature review (Agrawal et al., 2013; Chen et al., 2005b; Storr et al., 2011; Lynskey et al., 2012; O'Keefe et al., 2016; Sartor et al., 2015, 2013a; Behrendt et al., 2009; Chamla et al., 2006). The methodology can incorporate time-varying effects, whereby the status of certain factors can be modelled to alter over time. For example, if an individual develops an additional health problem during the period of time that is being analysed, this can be modelled so that the effect of this is not incorporated into the analysis until the time at which the problem occurred. This methodology allows for both the multi-stage nature of drug use, as the starting and ending time points can be set, and is designed to analyse and account for the complexities of time data.

Table 1.1: Results from Studies Utilising Two-Stage Twin Modelling in Drug Use

Paper	Drug	Stages of Use	Genetic correlation		
			Males	Females	Overall
Broms et al. 2006	Tobacco	Initiation and Amount smoked	.23	.17	-
		Initiation and Cessation	.22	-	-
Grant et al. 2009	Alcohol	Heaviness of Use and Abuse/Dependence	-	-	.97
Morley et al. 2007	Tobacco	Age Tobacco Onset and Cigarette Consumption	.82	.59	-
		Cigarette Consumption and Smoking Persistence	.88	.43	-
		Age Onset and Smoking Persistence	.23	.00	-
Pagan et al. 2006	Alcohol	Initiation and Frequency of use	-	-	.23
		Initiation and Problem Use	.29	.15	-
		Frequency of Use and Problem Use	.63	.78	-
McCutcheon et al. 2012	Alcohol	Alcohol Use Disorder (AUD) 2+ symptoms and Remission	-	-.33	-
		Alcohol Use Disorder (AUD) 3+ Symptoms and Remission	-	.29	-
Sartor et al. 2010	Cannabis	Number of Times of Use and Number Dependence Symptoms	-	-	.98
	Alcohol	Heaviness of Use and Number of Dependence Symptoms	-	-	.95

1.6.2 Genetically Informative Methods

Given consistent evidence that there are genetic influences acting on speed of transitions (Lynskey et al., 2010; Sartor et al., 2009a; Lynskey et al., 2006, 2003), there is value is utilising genetically informative methods in this thesis. Classical twin modelling can be used, as in previous studies providing estimates of the heritability of SUDs, age of opportunity and age of initiation of use. However, in order to determine whether the genetic factors affecting speed of transition overlap with those affecting SUDs a more nuanced analytic approach is required. Two-stage multivariate twin modelling allows estimation of the correlation between genetic influences on two behaviours in situations where an earlier behaviour, such as opportunity to use a drug, is *necessary* for the expression of later behaviours, such as the development of dependence (Heath et al., 2002). This issue applies in many studies of alcohol, tobacco and drug use (Neale et al., 2006a), and consequently this approach has been applied previously in research into tobacco, alcohol and cannabis (Broms et al., 2006; Grant et al., 2009; McCutcheon et al., 2012; Morley et al., 2007; Pagan et al., 2006; Sartor et al., 2010). A summary of the drugs previously studied using this method, and the findings regarding genetic correlation, are presented in Table 1.1. These studies have consistently demonstrated a genetic correlation between the earlier and later stages of drug use, although the magnitude of the effect differs. This methodology allows for the multi-stage nature of drug use, and for the study of the consistency of influences across drug use stages.

1.7 Thesis Aims

Based on the results of the literature review, and drawing on the identified gaps in the literature, the following aims will be addressed in this thesis.

Aim 1

Using a general population sample, explore the relationship between the speed of transitions to cannabis opportunity and initiation to subsequent use, and later cannabis daily use, abuse and/or dependence, and treatment-seeking.

Hypothesis 1

Individuals who have faster transitions in early cannabis use will have an increased likelihood of later cannabis daily use, abuse and/or dependence, and treatment-seeking.

Hypothesis 2

Associations between speed of transitions in cannabis and increased likelihood of later cannabis daily use, abuse and/or dependence, and treatment-seeking will persist after control for identified individual, childhood, mental health and other drug use confounding variables.

Aim 2

Using a general population sample, explore the extent to which the speed of transition to cannabis opportunity and from opportunity to dependence are influenced by individual, childhood, mental health and other drug use factors.

Hypothesis 1

There will be individual variation in the speed of the transitions studied.

Hypothesis 2

The associations between speed of transition in cannabis use and individual, childhood, mental health and other drug use factors will differ between the stages of cannabis use.

Aim 3

Examine the extent to which the speed of early stage transitions in trajectories of cannabis use are influenced by additive genetic, shared and non-shared environmental influences; explore the extent to which genetic influences on these transitions are unique to the phenotype, and the extent to which they are correlated with cannabis dependence.

Hypothesis 1

Opportunity to use cannabis and time from initiation to subsequent use will be influenced by additive genetic factors.

Hypothesis 2

Heritable influences on speed of early transitions will be highly correlated with heritable influences on the liability to cannabis dependence

Aim 4

In a clinical sample, explore the relationship between the speed of early heroin transitions and later heroin dependence severity, overdose, injecting behaviours and

heavy heroin use, and the effect of route of administration on speed of transition.

Hypothesis 1

Individuals who have faster early transitions in heroin use will have increased dependence severity at treatment seeking, time to treatment seeking, overdose, and injecting behaviour.

Hypothesis 2

Associations between speed of specific early transitions in heroin use and increased dependence severity at treatment seeking, time to treatment seeking, overdose, and injecting behaviour will persist after control for demographic factors.

Hypothesis 3

Injecting route of administration will be associated with faster speed of transition to daily heroin use.

CHAPTER 2

Methodology

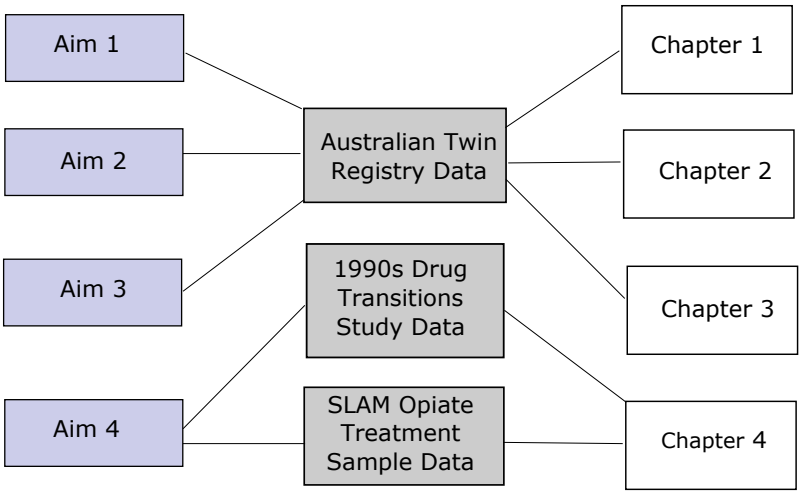
2.1 Rationale

The literature review (see Chapter 1) identified a number of gaps in the literature. The relationship between early transitions in drug use and important clinical outcomes has not been systematically studied, it is unknown whether factors influencing progression in drug use are consistent across stages of drug use, and there are novel transitions that have not been explored. By utilising variation in the speed of transitions in drug use and using a combination of epidemiological and genetically informative study designs, I address the aims of this thesis (see Chapter 1). An outline of the structure of the thesis is presented in Figure 2.1.

2.2 Sample 1: Australian Twin Study Of Cannabis And Other Illicit Drug Misuse

Chapters 3, 4 and 5 are based on secondary analysis of existing data from a cross-sectional survey of Australian born twins interviewed 2005-2009. The Australian Twin Study Of Cannabis And Other Illicit Drug Misuse (Lynskey et al., 2012) was established to collect detailed phenotypic information on patterns of cannabis use and cannabis-related symptomatology, associated use, abuse, and

Figure 2.1: Outline of Thesis Aims, Data Used, and Chapters



dependence on both licit and illicit drugs; psychiatric comorbidity including conduct disorder, major depressive disorder, social phobia, and panic disorder; child and family circumstances including opportunity to use drugs, exposure to inter-parental conflict, parenting practices, and peer affiliations. Using these data, I explore the relationship between speed of early transitions in cannabis use and later cannabis daily use, abuse/dependence and treatment-seeking, and to identify influences on the speed of transitions in cannabis use.

2.2.1 Procedure

The Australian Twin Registry

The participants included in this study were recruited via the Australian Twin Registry (ATR). Full details on the structure and processes of the ATR can be found in Hopper et al. (2013) and Hopper (2002). Established in 1981, the ATR is administered through the University of Melbourne and recruits twins from across Australia. At the time of data collection on the present study, an executive committee consisting of researchers with expertise in twin research, a gynaecologist specialising in multiple births, a representative of the Australian Multiple Births Association, and a twin representative, oversaw the ATR. A selection of this committee approved the Australian Twin Study Of Cannabis And Other Illicit Drug Misuse. In order to reduce attrition and ensure data quality, the ATR has

staff who manage on-going recruitment, participant information and contact with members of the registry. Each year these staff update the records of more than 7000 pairs (roughly a quarter of the members of the registry), keeping records of active members accurate.

The ATR is a voluntary registry. Twins are recruited through their own volition in adolescence or adulthood, and the ATR advertises through Association of Multiple Births clubs, word of mouth, schools, medical centres, posters, and electronic and print media. At the end of 2001, nearly 31000 pairs were registered with the ATR. Of those who had registered, 91%were "active" pairs, meaning that both members were alive, willing to participate and able to be traced by the ATR. When joining the ATR, participants complete baseline questionnaires on basic demographics, lifestyle and general health.

At most recent estimate, 22% of the ATR were monozygotic females, 16% monozygotic males, 17% dizygotic females, 15% dizygotic males, and 25% dizygotic opposite-sex pairs. There were also 5% same-sex twin pairs of unknown zygosity. The average age of members was 34 years (age range from 1 month to 99 years), with 24% under the age of 18 years (Hopper et al., 2013).

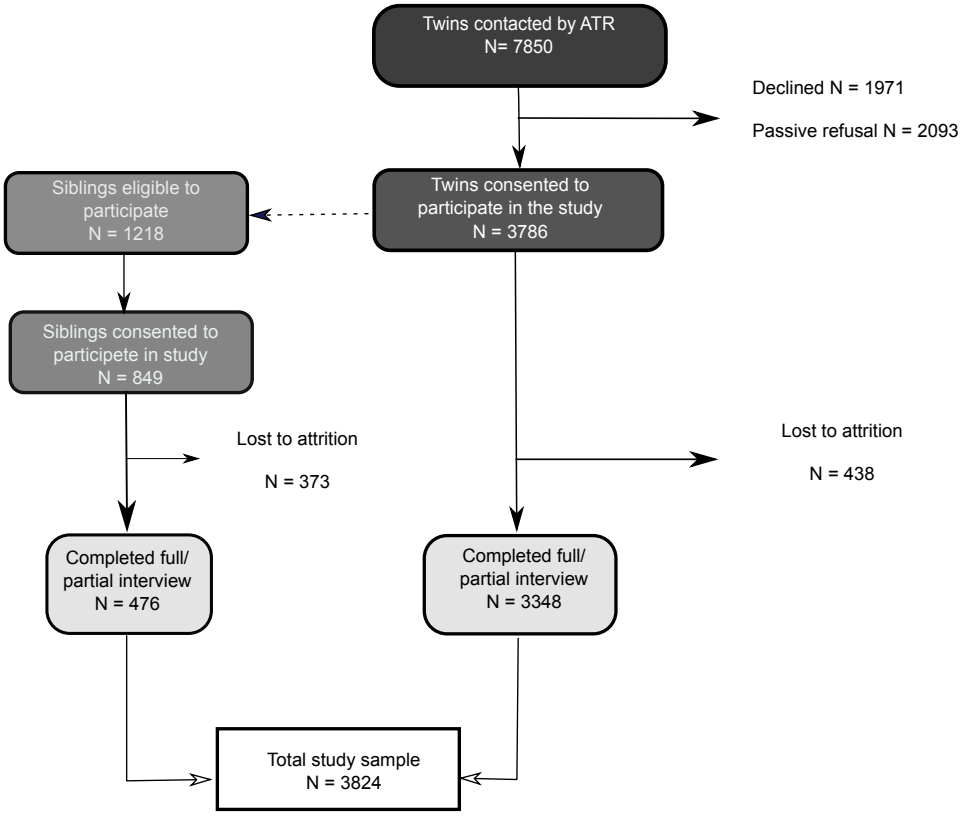
Recruitment and Data Collection

ATR pairs born 1972 - 1979 were reached through a two-tiered process, required by the ATR ethics committee. Twins initially received a letter from the ATR outlining the study and the review process that it underwent, and this was followed by telephone contact. Once twins had been provided with details of the study purpose, procedures involved, associated risks, feedback that would be provided, and who to contact for further information by the ATR, they were asked if they were willing to have their name and contact details forwarded to the team leading the study, who were based at Queensland Institute of Medical Research (QIMR).

Those researchers then contacted the twins who had agreed to provide their details, in order to explain the study purposes and enrol those who wanted to participate. Factors that may have affected recruitment in this study included the burden of time (which this study sought to compensate through financial reimbursement of a $50 (Australian Dollars) gift card on completion of the study), and concerns about privacy and confidentiality (Singer and Presser, 2007). Due to low response to ATR contact, the recruitment sample was extended to include

siblings of twins who had taken part to increase the power for the genetic design. A total of 3824 individuals participated in the interview, forming the complete sample for the analyses in this thesis (Lynskey et al., 2012). Figure 2.2 summarises the recruitment process and provides response rates at each stage of the data collection. The composition of the twin sample is provided in Table 2.1.

Figure 2.2: Outline of Recruitment to the Australian Twin Study Of Cannabis And Other Illicit Drug Misuse



Data were collected through Computer Assisted Telephone Interview (CATI) with a trained researcher. These researchers were lay interviewers who had received two weeks of structured interview training, and all interviews were recorded by audiotape (if permission was granted by the interviewee) in order to ensure data quality. Different interviewers conducted the interviews for each family member, in order to avoid any bias that could be introduced through the interviewer possessing prior knowledge of the twin or their family members. CATI methods allow for more complex questionnaires than those conducted using pen and paper, as computer scripts can provide tailored questions, and allow the interview to be

routed to ensure all relevant questions are asked (Hansen, 2007).

Table 2.1: Composition of the ATR Sample

Zygosity	Total N	N Pairs	N Unpaired twins
Monozygotic females	976	398	180
Monozygotic males	490	173	144
Dizygotic females	741	305	131
Dizygotic males	373	118	137
Dizygotic opposite sex	746	231	284
Unknown zygosity female	13	-	-
Unknown zygosity male	9	-	-
Siblings female	267	-	-
Siblings male	209	-	-

Funding and Ethical Approval

This research was funded by National Institute on Drug Abuse (NIDA) grant: DA18267 and facilitated through review by the Australian Twin Registry (as described above), a national resource supported by an Enabling Grant (ID 628911) from the National Health & Medical Research Council (Lynskey et al., 2012). Ethical approval for data collection was obtained from QIMR Berghofer Medical Research Institute and the Washington University School of Medicine. King's College London Research Ethics Subcommittee approved access and storage of the data.

The Semi-Structured Assessment for the Genetics of Alcoholism

The Semi-Structured Assessment for the Genetics of Alcoholism (SSAGA-II), a semi-structured measure of mental health, formed the basis of the CATI. This was developed for genetic studies of alcoholism, and was created to explore polydiagnosis, co-morbidity, and how phenotypes are expressed (Bucholz et al., 1994). The measure incorporates DSM-III, DSM-IV and ICD10 diagnostic criteria to allow comparison to other research, and non-clinical researchers who are trained for a week in advance can administer the measure.

The SSAGA-II has good reliability and validity. Interrater reliability for cannabis use disorders in cross-centre tests of all subjects had kappas of 0.72 for cannabis dependence, 0.63 for cannabis abuse and 0.82 for cannabis abuse or dependence. Within-centre tests showed similar results, with kappas of 0.76 for

cannabis dependence, 0.53 for cannabis abuse and 0.82 for cannabis abuse or dependence (Bucholz et al., 1994). To measure validity the SSAGA has been compared to the Schedule for Clinical Assessment in Neuropsychiatry (SCAN). For cannabis dependence sensitivity (proportion of true cases correctly identified) was 73.3%, and specificity (proportion of non-cases correctly identified) was 86.1% (Hesselbrock et al., 1999; Prince, 2003). The kappa for the cannabis dependence diagnosed on the SSAGA compared to the positive symptoms identified by the SCAN was 0.71. The version of the SSAGA used in the present study was the SSAGA-OZ, a version modified for telephone interview in an Australian population. Modification of telephone interview did not affect the test-retest reliability and accuracy of self-report (compared to family report) on this measure (Heath et al., 1997).

2.2.2 Measures

The CATI collected data on a number of drug use behaviours, and analyses presented in this thesis focus on cannabis use. A detailed description of items used and variables derived is provided below.

Cannabis Variables

Age of First Opportunity to Use Cannabis

To measure opportunity to use cannabis participants were asked “have you ever been offered, or had the opportunity to use marijuana/hash, even if you didn’t use them at the time?". Those reporting a lifetime opportunity to use cannabis were then asked: “How old were you the first time?”.

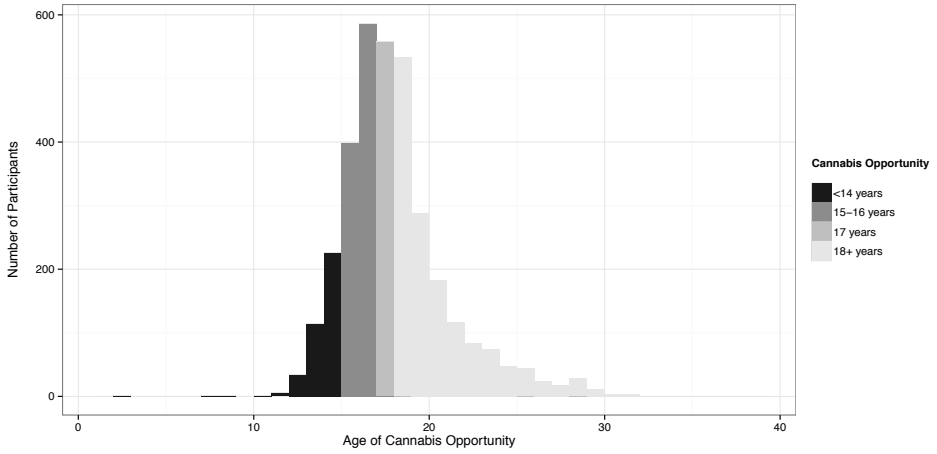
Table 2.2: Percentile groups for calculating analysis groups for speed of transition to opportunity to use cannabis

Percentile	Maximum age	Age range	Analysis group N(%)
10th	14	2-14	388 (11.4)
25th	16	15-16	986 (29.0)
50th	17	17	588 (16.4)
Reference category	38	18-38	1467 (43.2)

Out of a total of 3824 interviewed participants, data were available for 3798 participants. Of these, 89.5% (N=3399) reported having a lifetime opportunity to use cannabis.

For categorical analyses on speed of transition to cannabis use opportunity the 10th, 25th, 50th percentiles of the age of onset distribution were used to determine transition speed groups (see Table 2.2). This formed a four level categorical variable with the groups' opportunity age 14 and under, opportunity age 15-16, opportunity age 17, and opportunity age 18 and over, which was used for all analyses in Chapter3. See Figure 2.3 for distribution, and age categories used in analysis.

Figure 2.3: Distribution of age of opportunity to use cannabis



Cannabis Use To measure lifetime cannabis use participants were asked "have you ever used either of [marijuana or hashish]?". Those reporting lifetime cannabis use were then asked: "How old were you the first time you used [marijuana or hashish]?". Out of a total of 3824 interviewed participants, data were available for 3797 participants. Of these, 68.5% (N=2601) reported lifetime cannabis use. Reporting on opportunity to use cannabis was not required for participants to report lifetime cannabis use.

There is no consensus in the literature as to what constitutes early onset, which has been categorized as anywhere between 11 – 18 years, but in line with a number of existing publications (Agrawal et al., 2006; Michael T. Lynskey et al., 2012; Sartor et al., 2009) the selected cut-off for early onset was those who were aged 16 and

under when cannabis was first used. Early onset of cannabis use was reported by 26.33% (N=1007) of participants.

Time to Subsequent Use of Cannabis

Those who reported using cannabis more than once were asked: "how soon after you first tried marijuana did you try it again?" Responses to this question were collected using the following response options:

- The same day
- The next day
- Within a week
- Within a month
- Within 3 months
- Within 6 months
- Within a year
- More than a year

Progression to subsequent cannabis use was reported by 59% (N=2239) of participants, and 86% of those who reported ever using cannabis.

Daily Cannabis Use

Participants who reported using cannabis more than 11 times were asked if they had used cannabis at least once a day during their period of heaviest cannabis use. Lifetime daily cannabis use was reported by 9.75% (N=373) of participants, and 16.7% of those who reported ever using cannabis.

Cannabis Abuse

Participants were classified as meeting DSM-IV criteria (American Psychiatric Association, 2000) for lifetime cannabis abuse if they reported one or more of the following within a 12 month period:

- Often using cannabis in a situation where they might get hurt
- Arrested more than twice within a 12 month period as a result of their cannabis use
- Cannabis use having caused difficulty with work, study or household responsibilities
- Cannabis having caused social and interpersonal problems

Lifetime cannabis abuse was reported by 15.3% (N=581) of the whole sample, and 22.3% of those who reported lifetime cannabis use.

Cannabis Dependence

Participants were classified as meeting lifetime criteria for DSM-IV (American Psychiatric Association, 2000) cannabis dependence if they reported 3 or more of the following symptoms occurring within the same 12 month period:

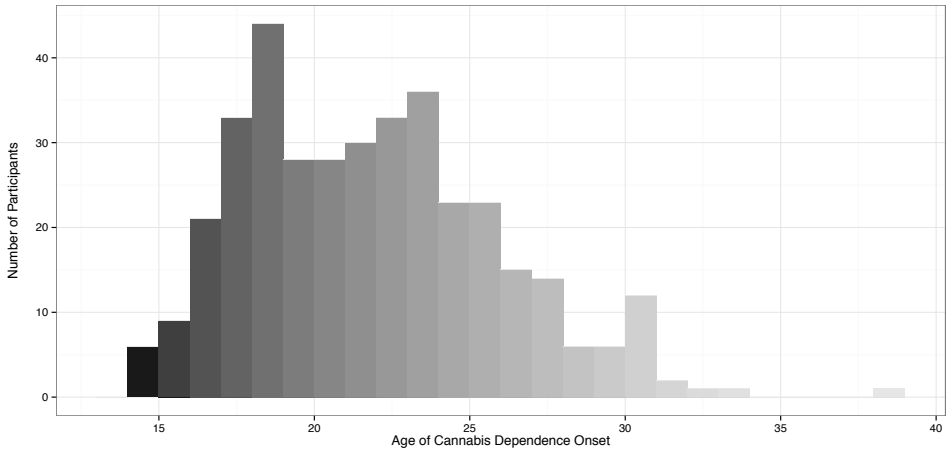
- Using cannabis a greater number of times/greater amount than was intended
- Tolerance
- Wanting to cut down/stop use
- Spending so much time obtaining/using/recovering from the effects of cannabis that the participant had little time for anything else
- Reducing important activities as a result of cannabis use
- Continuing use despite it worsening health/emotional problems

Withdrawal was not included as it was not part of DSM-IV criteria for cannabis dependence.

The mean age of dependence onset was 21.4 (s.d. 4.1, range 14-38). Lifetime cannabis dependence was reported by 9.73% (N=372) of participants, and 14.5% of those who had ever reported using cannabis.

Age of onset of cannabis dependence was ascertained by asking participants how old they were the first time they experienced three or more of these symptoms within the same 12 month period. See Figure 2.4 for distribution.

Figure 2.4: Distribution of age of cannabis dependence onset



Lifetime Cannabis Abuse and Dependence

For analyses examining associations between early transitions and later cannabis

outcomes, meeting cannabis abuse criteria and meeting cannabis dependence criteria (outlined above) were combined into a composite measure of abuse and/or dependence. Lifetime cannabis abuse and/or dependence was reported by 16.32% (N=624) of participants, and 24.1% of those who reported ever using cannabis.

Treatment-Seeking for Cannabis Use

Participants self-reported seeking treatment for cannabis problems from a professional and were asked to describe who they had sought treatment from: Psychiatrist (N=45), General Practitioner or other medical doctor (N=80), Psychologist (N=42), another mental health professional (N=61), member of the clergy (N=7) or another source (N= 9). Seeking treatment for cannabis use was reported by 3.5% of the whole sample (N=133), and 5.9% of those who had reported lifetime cannabis use.

Demographic Covariates

Sex/Gender

Sex/gender was determined through self-report. 63.7% of the sample was female.

Education

Participants were asked to report their highest level of education from the options primary incomplete, primary complete, year 8 completed, year 9 completed, year 10 completed, year 11 completed, year 12 completed, Technical and Further Education college, undergraduate degree, or post-graduate degree. For analysis, respondents were classified by whether or not their highest level of education was post-secondary/higher education. Completing post-secondary/higher education was reported by 24.6% (N=941) of participants.

Childhood Environment Age 6-13

Single Parent Family

Interviewers recorded whether participants lived with both their mother/mother figure and their father/father figure for at least 4 full years between ages 6-13. Growing up in a single parent family was reported by 5.75% (N=220) of participants.

Parental Conflict

Respondents were asked “How often did your parents fight or argue in front of you” and “How much conflict and tension was there between your parents”. Participants

who reported parents 'sometimes' or 'always' fought or argued, or reported 'a lot' or 'some' conflict/tension were coded as experiencing high parental conflict. High parental conflict was reported by 36.85% (N=1409) of participants.

Strict Parenting

Respondent were asked "In your opinion, when you were 6 to 13, was your mother/mother figure more strict than most mothers?" and " In your opinion, when you were 6 to 13, was your father/father figure more strict than most fathers?". Responses were either yes or no. Those who endorsed either of these items were classified as having experienced strict parenting (N=1865, 48.9%).

Childhood religious attendance

Childhood religious service attendance was determined through participant self-report to the question "how often did you attend religious services between the ages of 6 and 13?". Participants selected from the options more than once a week, once a week, once or twice a month, every couple of months, once or twice a year, rarely, or never. Participants were coded as regularly attending religious services if they reported attendance more than once a week, once a week, once or twice a month or every couple of months. Regular religious attendance was reported by 60.02% (N=2295) of participants.

Childhood Adversity

Childhood sexual abuse (CSA)

Individuals were asked "before age 18, were you ever forced into sexual intercourse or any other sexual activity?" Those who reported that they had were classified as having experienced CSA. Participants were then asked for the age at which they were first forced into sexual activity. Individuals who had reported an age of 18 or over for being forced into sexual activity (N=8) were recoded to missing, to account for the inconsistency in their response to the two questions. Childhood sexual abuse was reported by 8.3% (N=312) of participants. Mean age of onset of sexual abuse was age 10.9 (s.d. 4.6, range 0-17).

Parent Covariates

Parental alcohol problems

Participants were asked "Did drinking ever cause [your biological father/mother]

to have problems with health, family, job or police, or other problems?" and "Did you ever feel that [your biological father/mother] were excessive drinkers?" These measures were derived from the Family History Assessment Module (FHAM), which has been shown to have good accuracy at identifying alcohol dependence from family interviews (71.5% of cases were identified by at least one positive report, and 85% of non-cases had no positive reports; Rice et al. (1995)). Individuals who responded 'yes' to either of these questions were coded as experiencing parental alcohol problems (24.95% (N=954) of participants).

Parental drug problems

Participants were asked "Did using drugs ever cause [your biological father/mother] to have problems with health, family, job or police, or other problems?" and "Did you ever feel that [your biological father/mother] had a problem with drugs?" As with parental alcohol problems, these measures were derived from the FHAM (Rice et al., 1995). Individuals who responded 'yes' to either of these questions, were classified as having a parental history of drug problems. Parental drug problems were reported by 3.43% (N=131) of participants.

Peer Cannabis Use

High-School Peer Cannabis Use

The extent of cannabis use amongst high school peers was measured through self-report questions asking whether 'hardly any', 'some', 'half', 'three quarters' or 'almost all' the students who were in their grade in high-school used cannabis. Participants were categorised as being exposed to high levels of illicit drug use during high school if they reported that at least three quarters of their peers used cannabis. High levels of high school peer cannabis use were reported by 6.38% (N=244) of participants.

Mental Health Covariates

Conduct Disorder (CD)

Participants were coded as meeting criteria for CD if they reported at least 3 of the 15 DSM-IV criteria (American Psychiatric Association, 2000) occurring within the same 12-month period, and specified to refer to the time period before age 18:

- Aggression to people and animals

1. often bullies, threatens, or intimidates others
2. often initiates physical fights
3. has used a weapon that can cause serious physical harm to others (e.g., a bat, brick, broken bottle, knife, gun)
4. has been physically cruel to people
5. has been physically cruel to animals
6. has stolen while confronting a victim (e.g., mugging, purse snatching, extortion, armed robbery)
7. has forced someone into sexual activity

- Destruction of property
    1. has deliberately engaged in fire setting with the intention of causing serious damage
    2. has deliberately destroyed others' property (other than by fire setting)
- Deceitfulness or theft
    1. has broken into someone else's house, building, or car
    2. often lies to obtain goods or favours or to avoid obligations (i.e., "cons" others)
    3. has stolen items of nontrivial value without confronting a victim (e.g., shoplifting, but without breaking and entering; forgery)
- Serious violations of rules
    1. often stays out at night despite parental prohibitions, beginning before age 13 years
    2. has run away from home overnight at least twice while living in parental or parental surrogate home (or once without returning for a lengthy period)
    3. is often truant from school, beginning before age 13 years

Those who reported symptom onset after age 18 (N=5) were coded as missing, to account for the inconsistency in their response to the two questions. Conduct disorder criteria were met by 8.4% (N=320) of participants, and the mean age of

reported onset was 14.1 (s.d. 2.3, range 5-17).

Non-Clinical Depressive Episodes

A non-clinical depressive episode was recorded if participants reported a two week period during which they were more irritable than usual (if under age 18 at the time, in line with American Psychiatric Association (2000) DSM-IV guidance), felt depressed/down/sad/blue/discouraged, or had a lot less interest in things. Non-clinical depressive episodes were reported by 47.67% (N=1823) of participants.

Alcohol, Tobacco and Other Drug Use

Monthly Alcohol Use

Monthly alcohol use was measured through the interview item "At what age did you start to drink regularly - that is, drinking at least once a month for 6 months or more?" Lifetime regular alcohol use was reported by 90.72% (N=3469) of participants. The mean age of monthly alcohol use onset was 18.2 (s.d. 2.75, range 9-41).

Alcohol dependence

Participants were classified as experiencing lifetime alcohol dependence (American Psychiatric Association, 2000) if they reported 3 or more of the following DSM-IV criteria occurring within a 12 month period:

- Increased tolerance to alcohol use
- Drinking on occasions when did not intend to, or for a longer time than intended, or a greater amount than intended
- Wanted to or tried to cut down/quit, but found they could not
- Had a period of several days when drinking or recovering from the effects of alcohol meant there was little time for anything else
- Gave up or reduced important activities to drink
- Continued to drink alcohol despite causing/worsening health problems, having a negative effect on mental health, or experiencing blackouts
- Experienced withdrawal from alcohol

Lifetime alcohol dependence was reported by 24.82% (N=949) of participants. The mean age of alcohol dependence onset was 22.5 (s.d. 4.2, range 13 - 40).

Weekly Tobacco Use

Weekly tobacco use was measured through the interview item "Was there ever a

time in your life when you smoked cigarettes at least once a week for at least two months in a row?" Lifetime weekly tobacco use was reported by 40.5% (N=1545) of participants. The mean age of weekly tobacco use onset was 17.3 (s.d. 3.4, range 7 – 33).

Tobacco Dependence

Those sample members who reported smoking 100+ cigarettes in their lifetime were asked a series of further questions about their experiences of nicotine dependence symptomatology. Participants were classified as experiencing lifetime tobacco dependence (American Psychiatric Association, 2000) if they reported 3 or more of the following DSM-IV criteria occurring within a 12 month period:

- Smoking more than 20 cigarettes a day, or smoking more/using a stronger type of tobacco than when started smoking
- Often chain smoked cigarettes
- Gave up important activities/missed socialising due to being unable to smoke cigarettes
- Smoked more cigarettes than intended, or broke own rules on smoking
- Wanted to or tried to cut down/quit, but found they could not
- Experienced withdrawal from cigarettes
- Continued to smoke cigarettes despite causing/worsening health problems and anxieties

Lifetime tobacco dependence was reported by 25.52% (N=976) of participants. The mean age of tobacco dependence onset was 21.9 (s.d. 4.5, range 12 – 43).

Other Drug Use

Other drug use was recorded if participants reported lifetime non-prescribed use of any of the following:

- cocaine (all forms)
- stimulants
- opiates and major painkillers
- sedatives
- hallucinogens
- solvents and inhalants
- dissociative drugs

Lifetime illicit drug use was reported by 43.75% (N=1673) of participants. The mean age of other illicit drug use onset was 21.7 (s.d. 4.3, range 0 – 43).

Other Drug Dependence

Participants were classified as meeting criteria for lifetime other drug dependence (American Psychiatric Association, 2000) if they reported 3 or more DSM-IV criteria for dependence occurring within the same 12 month period for any one of the drugs listed above:

- Using the drug a greater number of times/greater amount than was intended
- Tolerance
- Wanting to cut down/stop use but being unable to
- Spending so much time obtaining/using/recovering from the effects of the drug that the participant had little time for anything else
- Reducing important activities as a result of drug use
- Continuing use despite it worsening health/emotional problems
- Experiencing withdrawal

All symptoms had to occur in relation to the same drug for dependence to be recorded. Lifetime other illicit drug dependence was reported by 4.68% (N=179) of participants. The mean age of onset was 23.0 (s.d 4.5, range 13 – 34).
Prevalence of dependence on individual drugs was:

- cocaine (all forms): 0.4%
- stimulants: 3.7%
- opiates and major painkillers: 0.9%
- sedatives: 0.7%
- hallucinogens: 0.3%
- solvents and inhalants: 0%
- dissociative drugs: 0.1%

Equality of Environment

The following self-reported items were asked only to twin pairs, to assess twin similarity: “When you were 6 to 13, how often did you share the same friends?”, “How often did you dress alike?”, "In primary school, how often were you in the same classes?”, “In high school how often were you in the same classes?”, “Between

the ages of 6 and 13, how often did your mother/ father/teachers/strangers have trouble telling you and your twin apart?", and "Without using a list, when you and your twin were children, were you as alike as 'two peas in a pod', or only of normal family physical likeness?". Concordance on these items was used to test the equality of the twin childhood environment.

2.2.3 Missing Data

Non-random patterns of missing data can bias results. As cannabis use and outcome phenotypes are the independent and dependent variables in the analyses on these data, analyses of patterns of missing data have been conducted on these variables.

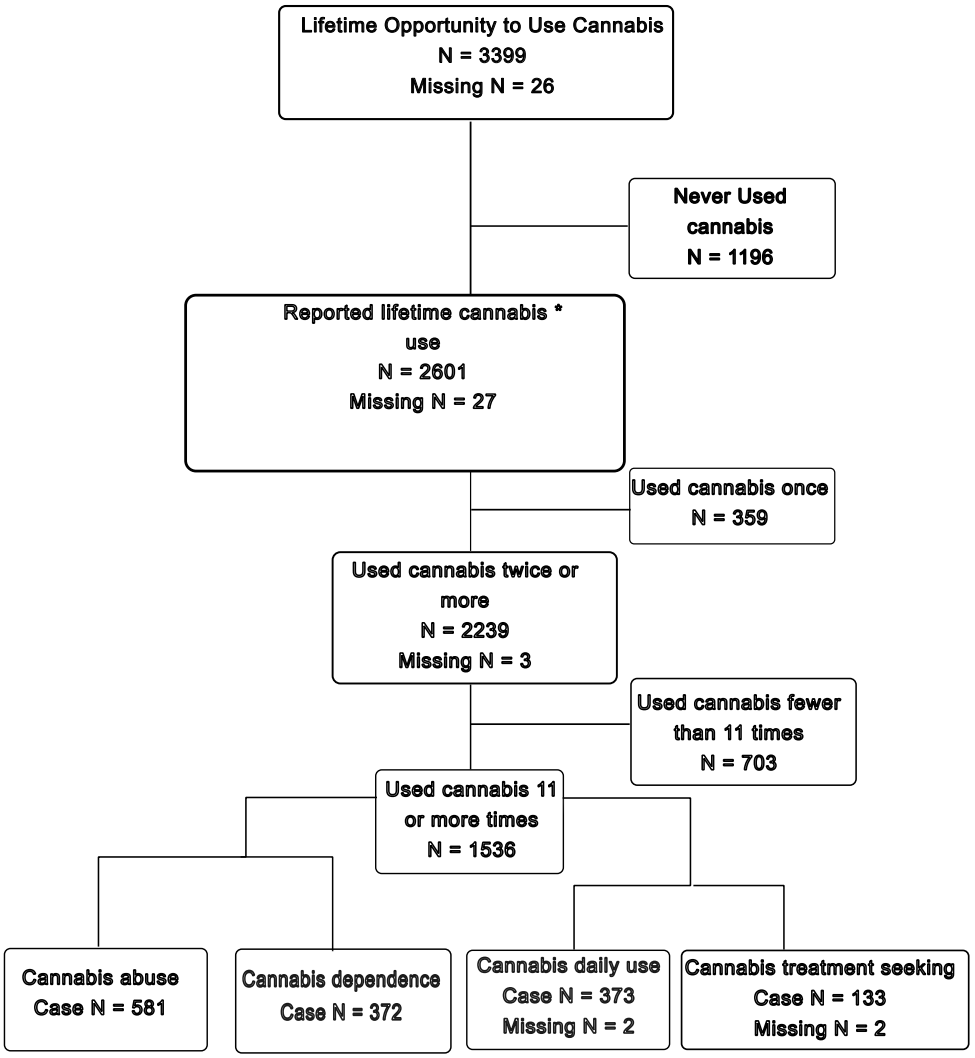
Due to the CATI method of data collection, there is very little missing data in this dataset. A full outline of the missing data for analysis variables can be found in Figure 2.5. This figure summarises the structure of the interview and provides details of both the number of respondents reporting a specific behaviour and the number of participants for whom there was missing data. A full list of the missing data for each covariate used in the analyses, and missing data on ages of onset where relevant to the analyses, can be found in Appendix 2.

For the majority of independent and dependent variables used within the analyses, the numbers of missing participants are so low ($N \leq 4$) that they did not introduce risk of bias. For opportunity to use cannabis and lifetime use of cannabis, missing and non-missing participants were compared on the demographic characteristics of gender and education using χ^2, and age at interview using anova tests. Statistical significance was determined at the $P \leq 0.05$ level.

Missing on Opportunity to Use Cannabis

Compared to those who responded to the item on opportunity to use cannabis (N=3798), those who were missing (N=26) were significantly different on both gender (36.1% of non-missing participants were male, compared to 65.4% of missing participants, P=0.002, OR 0.29, 95% CI 0.12 - 0.71) and education (24.4% of non-missing participants had completed further/higher education, compared to 56.5% of missing participants, $P \leq 0.001$, OR 4.0, 95% CI 1.76 -9.20). Mean age at interview was 32.1 for non-missing participants (S.D 3.0) and 32.4 for missing

Figure 2.5: Group Numbers and Missing Data Numbers for Cannabis Variables



participants (S.D 3.5) (F=0.18, df = 1, P=0.67). To account for the significant differences between missing and non-missing groups, analyses were adjusted for gender and education.

Missing on Lifetime Cannabis Use

Compared to those who responded to the item on lifetime cannabis use (N=3797), those who were missing (N=27) were significantly different on both gender (36.1% of non-missing participants were male, compared to 63.0% of missing participants, P=0.004, OR 0.33, 95% CI 0.14 - 0.77) and education (24.4% of non-

missing participants had completed further/higher education, compared to 54.2% of missing participants, P=0.001, OR 3.65, 95% CI 1.50 - 9.04). Mean age at interview was 32.1 for non-missing participants (S.D 3.0) and 32.4 for missing participants (S.D 3.5) (F=0.22, df = 1, P=0.30). To account for the significant differences between missing and non-missing groups, analyses were adjusted for gender and education.

2.3 Sample 2: South London and Maudsley NHS Trust (SLAM) Opiate Substitution Treatment Sample

This exploratory study was conducted in order to test hypotheses relating to speed of early heroin transitions and later outcomes, and to establish whether findings relating to speed of transition for cannabis use would also be found for a different drug class. Given the low prevalence of heroin use in the general population (see Chapter 1), a clinical population was required to study heroin use. Data collection took place in two Drug and Alcohol Treatment services within the South London and Maudsley NHS Trust (SLAM). The data from this study are used in Chapter 6.

2.3.1 Procedure

Service User Involvement

Participants in the Service User's Research Group at the Aurora Project, Lambeth, were invited to a focus group to discuss the study purpose and design. A number of individuals in recovery from drug and alcohol abuse attended the session, read and discussed the interview items and patient information, and provided feedback. As a result of this session, the following suggestions were incorporated into the study design:

- The interview was administered by a researcher as opposed to being produced as a questionnaire for participants to complete themselves, in order to avoid literacy issues.
- Following concerns raised by focus group participants that the proposed measures of severity of dependence (Severity of Dependence Scale) would not have much variation in the sample, additional items were included to

represent severity in the sample, including injecting behaviours and amount of heroin used.

- It had been proposed to use daily heroin use as a marker of having developed problem heroin use. Feedback strongly indicated that judgement of the onset of problem heroin use should not rely solely on the measure of daily heroin use, but should also incorporate measures such as effect on relationships and employment. There were concerns that daily heroin use may not be the first experienced sign of problems.

Recruitment and Data Collection

In total 93 participants were recruited into the study and 11 approached participants declined to participate, providing a participant response rate of 89%. Potential participants attending for opiate substitution treatment, who had capacity to take part, were identified and informed of the study by clinic staff. Interested participants contacted the PhD researcher located in the clinic waiting area, and clinic staff provided the researcher with refusal figures. The researcher accompanied the participant to a private room within the clinic, then outlined the study using the participant information sheet (see Appendix 4), and obtained informed consent from the participant (see Appendix 4). Any participants who were intoxicated, assessed by the researcher asking a screening question on intoxication, were asked to participate in the study at a later date.
Participants provided responses to a short structured quantitative interview administered by the researcher. This included demographic information, age of onset for a number of drug behaviours, and the Severity of Dependence Scale (see measure outline below; see Appendix 4 for full interview schedule). Participants were reimbursed £10 upon completion of the interview.

Sample Size and Power Calculation

The pre-planned primary analysis for this study was a regression model testing for an association between speed of early transition in heroin use and dependence severity. Assuming a positive linear relationship between speed of transitions in heroin use and dependence severity, power analysis was based on whether

the standardized regression co-efficient or slope differed from 0. Power was calculated in G-Power 3.1 (Faul et al., 2007) using a linear bivariate model. With 93 participants and assuming an alpha level of 0.05 the study has 91% power to detect a standardized regression slope of 0.3.

Data security

Participants were assigned an anonymous study identification number, including a clinic location identifier. Completed interviews were kept separate from consent forms at all times. Completed interviews were kept in a locked bag when in the clinic and when transporting to the Institute of Psychiatry, Psychology and Neuroscience (IoPPN). On arrival at the IoPPN, records of completed interviews were stored in a secured cabinet. Completed consent forms were stored in a separate but equally secure locked cabinet on a different floor. When responses were entered onto the computer for analysis, the data were stored on a secure IoPPN server that was only accessible to the research team.

Data entry

Data were double-entered using a custom designed spreadsheet in Microsoft Excel in preparation for data analysis. In order to minimise entry inaccuracies self-validating drop-down boxes containing the interview response options were created for each categorical item. Any inconsistent entries were checked against the original interviews and corrected. The double entered and checked data was then transferred into a Stata file (StataCorp, 2009) for analysis.

Ethical approval

Ethical approval was granted by the NHS Research Ethics Committee (15/LO/0705) and SLAM Research and Development Department.

2.3.2 Measures

The following analysis items were derived from the interview data. The full interview can be found in Appendix 4.

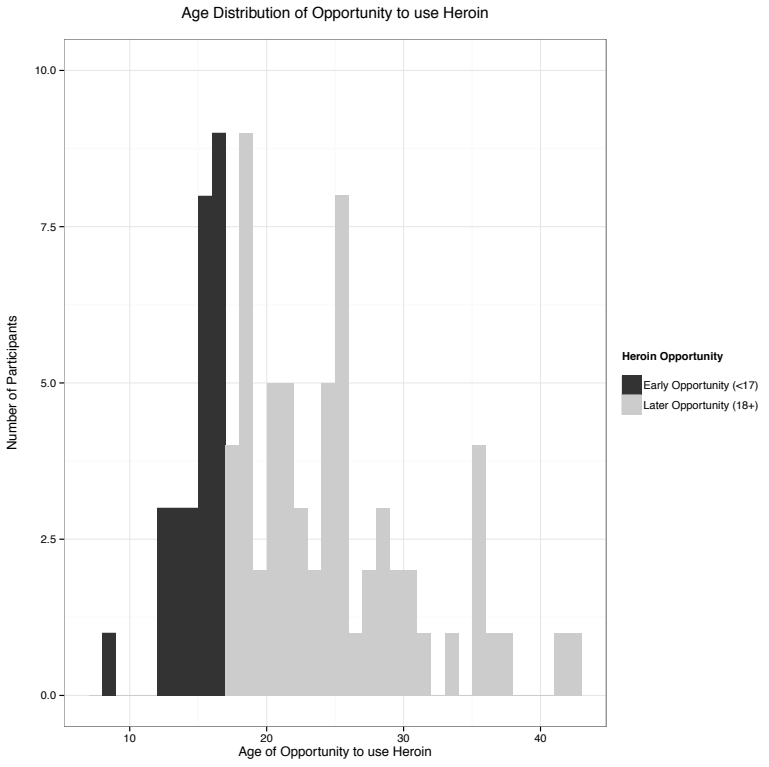
Speed of Transition Variables

Early Opportunity to Use Heroin

Participants self-reported their age at first opportunity to use heroin. To aid recall and improve understanding, participants were given the following prompt: "By an opportunity I mean someone either offered you heroin, or you were present when others were using and you could have used if you wanted to" (definition from Storr et al. (2011)).

As there is no known precedent in the literature for what is considered early opportunity to use heroin, the continuous age measure of this item was transformed into tertiles. Participants were in the lowest tertile for reported age of opportunity if they had opportunity to use heroin at age 17 or under. A binary variable of early/later opportunity was created using this data. In the sample 37.6% (N=35) reported early opportunity to use heroin. See Figure 2.6 for distribution.

Figure 2.6: Distribution of age of opportunity to use heroin



Time From Opportunity to Use Heroin to Initiation of Use

Participants self-reported the time between their first opportunity to use heroin and

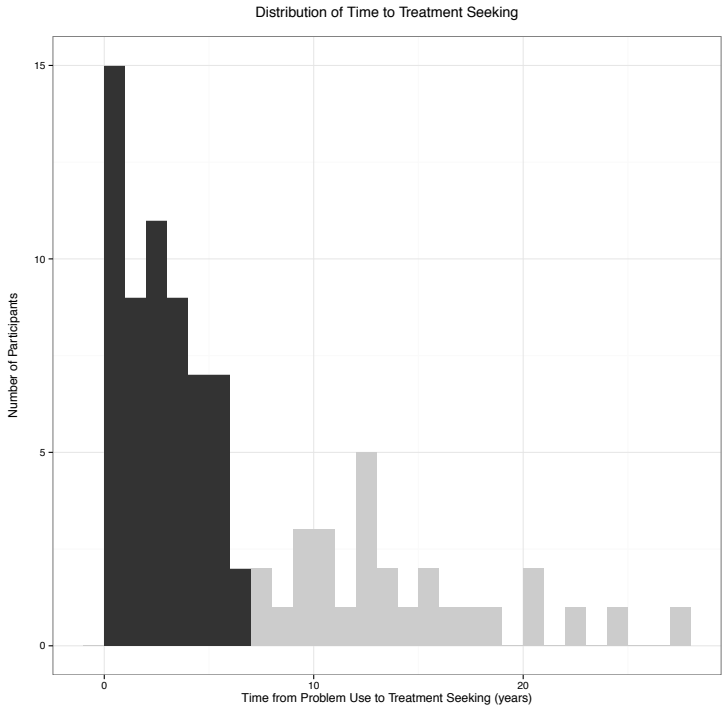
initiation of heroin use. Participants provided responses in days, weeks, months or years. These responses were coded to a dichotomous variable of "within a week of opportunity" and "more than one week after opportunity". In the sample 64.7% (N=57) reported progressing to initiation of use within a week of opportunity to use heroin.

Time From Initiation to Subsequent Heroin Use

Participants self-reported the time between their initiation of heroin use to their subsequent use of heroin (clarified in the interviewer as "the second time in your life that you used heroin"). Participants provided responses in days, weeks, months or years. These responses were coded to a dichotomous variable of "within a week of initiation" and "more than one week after initiation". In the sample 50.4% (N=47) reported using heroin for a second time within a week of initiating use.

Outcome Variables

Figure 2.7: Distribution of Time from Heroin Problem Use to Treatment Seeking



Time From Problem Use Onset to First Seeking Treatment

Participants' self-report of age at onset of problem heroin use, ascertained through

one or more of daily heroin use, using heroin alone, experiencing withdrawal symptoms, experiencing relationship problems as a result of heroin use, and experiencing employment/studying problems as a result of heroin use was utilised to create this variable. Mean age of first reported problem was 24 (s.d. 7.9, range 8 – 49). The earliest reported age of any of these was taken as the age of first reported problem with heroin use. Mean age at first treatment-seeking was 29.4 (s.d. 8.4, range 15 - 51). The time from the onset of problem use to treatment-seeking was calculated by selecting the age of first problem onset, and calculating the difference between this and the age at treatment-seeking. Any negative values (indicating that treatment-seeking was initiated before onset of any of these problems) were recoded to missing (N=3). The mean time from first problem to first seeking treatment was 5.9 years (s.d. 6.3, range 0 - 27 years). See Figure 2.7 for distribution.

Severity of Dependence at Time of Treatment-Seeking

The Severity of Dependence Scale (SDS) was administered as part of the interview. The SDS is a validated unidimensional measure (González-Saiz et al., 2008; Gossop et al., 1995) that consists of five items investigating drug taking behaviours that are related to dependence severity (Gossop et al., 1995) and measures the degree to which users are experiencing psychological dependence through a scoring system, with a score of 4 indicating dependence and higher scores indicating greater degrees of dependence severity (Ding et al., 2013; González-Sáiz et al., 2009).

The SDS has previously been tested for validity in London heroin using populations, and has positive correlations with heroin dose ($r=0.24$, $P\leq0.001$), duration of heroin use ($r=0.27$, $P\leq0.001$) and frequency of use in days per week ($r=0.43$, $P\leq0.001$), and participants who were using heroin daily, spending the majority of time with other users and being in contact with treatment services had significantly higher SDS scores (Gossop et al., 1995). Amongst an Australian methadone treatment sample SDS scores were positively correlated with current use ($r=0.44$, $P\leq0.001$), frequency of use of heroin ($r=0.19$; $P\leq0.001$, respectively) and duration of heroin use ($r=0.34$, $P\leq0.001$), and negatively correlated with time in methadone treatment ($r=-0.37$, $P\leq0.001$) and methadone dose ($r=-0.20$, $P\leq0.05$) (Gossop et al., 1995). Principal components analysis has demonstrated that the items within the SDS load onto a single factor, with good internal consistency (Cronbachs Alpha 0.8 – 0.9) observed in the tested heroin-using samples (Gossop et al., 1995).

The SDS is the only measure of dependence severity to include items focussing on compulsive drug taking and seeking (Conway et al., 2010), and has previously been shown to be appropriate for determining variation in dependence in opiate treatment populations (Gossop et al., 1995; Miller et al., 2014). The SDS items are scored never/almost never, sometimes, often or nearly always/always for the items:

- Do you think your use of opiates is out of control?
- Does the prospect of missing a fix (or dose) or not chasing make you anxious or worried?
- Do you worry about your use of opiates?
- Do you wish you could stop using opiates?

and scored not difficult, quite difficult, very difficult and impossible for the item:

- How difficult do you find it to stop or go without opiates?

Severity of opiate dependence at time of initial treatment-seeking was assessed through retrospective participant SDS self-report. Total scores were used for the analysis, with higher scores indicating increased dependence severity. The mean score at treatment-seeking was 10.2, s.d. 4.1, range 0 – 15. See figure 2.8 for distribution.

Injecting into the Groin or Neck

Participants self-reported whether they had ever engaged in femoral (groin vein) injecting or neck injecting as a result of their heroin use. These two individual items were combined into one measure of lifetime groin or neck injecting. In the sample, 44.1% (N=41) reported injecting into the groin or neck.

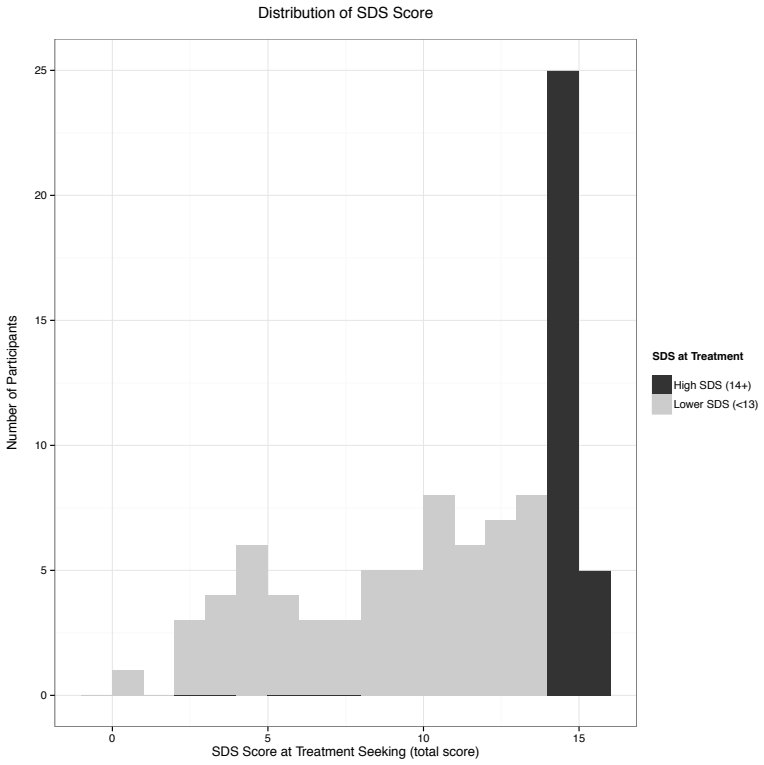
Overdose

Participants self-reported whether they had ever experienced overdose as a result of their heroin use. In the sample, 54.8% (N=51) reported experiencing overdose.

Heavy Use of Heroin

Information on the amount of heroin used was collected through the item "At your period of heaviest use, what amount of heroin/opiates were you using on a typical day?". Participants were permitted to respond with an amount in grams and/or in pounds sterling (£). The continuous measures of these responses were transformed into tertiles. Participants who reported being in the highest tertile for

Figure 2.8: Distribution of Heroin Severity of Dependence Score



reported amount in grams or in £ were classified as heavy users of heroin. In the sample, 36.4% (N=32) reported heavy heroin use.

Demographic Covariates

Gender

Gender was self-reported by participants: 75% (N=69) were male.

Ethnicity

Participants were asked to classify their ethnicity from the options: White British, White Irish, Other White, White and Black Caribbean, White and Black African, White and Asian, Other Mixed, Indian, Pakistani, Bangladeshi, Other Asian, Caribbean, African, Other Black, Chinese, Other. The variable was collapsed into the categories White British (55.9%, N =52), Other White (22.6%, N=21), and Non-White (21.5%, N = 20).

Age at Interview

Participants self reported their age at the time of the interview. The mean age was 42.1 (s.d. 7.4, range 22 – 63).

2.3.3 Missing Data

The only speed of transition variables missing data were 'age of opportunity to use heroin' (missing N=3) and 'time from opportunity to use heroin to initiation of use' (missing N=5). The outcomes with missing data were 'heavy opiate use' (missing N=5), and 'age at treatment-seeking for opiate use' (missing N=2), which would contribute to the variable of 'time from problem use to treatment-seeking'. These numbers were considered low enough for missing data to present no concerns. Information on missing data for all independent variables, dependent variables and covariates is available in Appendix 2.

2.4 Sample 3: The 1990s Drug Transitions Study

Chapter 6 includes analysis of data from a study conducted in the early 1990s in London (Griffiths et al., 1994). This study aimed to explore the nature and extent of transitions in route of heroin administration, using a sample that consisted of heroin users both in and out of treatment who had a variety of heroin administration routes. Prior to 1980, the predominant route of administration among London-based heroin users was injecting, but after 1980 chasing (inhaling heroin vapour) became more prevalent as a route of administration (Strang et al., 1992). Thus, at the time the study was conducted, the population contained individuals who had initiated heroin use by both routes. Utilising a population with variation in the route of heroin administration allows for specific testing of the relationship between Initial Heroin Administration Route (IAR) and transition speed.

2.4.1 Procedure

Recruitment and Data Collection

The sample consisted of 408 heroin users who were recruited in the London area during 1991. All participants had used heroin in the month prior to interview. The

sample was structured to include 200 subjects in contact with treatment agencies and 200 subjects who were not in treatment. Participants were defined as in treatment if they were attending a drug dependence unit, attending voluntary or statutory street agencies, in contact with a general practitioner specifically for a drugs problem, or in contact with any other organization specifically for a drugs problem.

Recruitment and interviewing were conducted by Privileged Access Interviewers (PAIs) (Griffiths et al., 1993). This method utilises individuals who have attributes that make them non-threatening to members of the population under study, and who have access to populations that traditional researchers would find difficult to reach. Current drug use was neither an inclusionary nor exclusionary criterion for the PAIs, who were selected based on:

- Having existing contacts (or the ability to easily develop contacts) within the population of interest
- Having personal attributes and life experiences that made them non-threatening to the population
- Being socially and educationally equipped to conduct the interview schedule, and having a relatively stable lifestyle that allowed employment
- conducting the interview and making contacts not being damaging to the PAI.

The mean number of interviews conducted by each interviewer was 22. Methodologies such as PAI can have a number of limitations, including being labour intensive, the risk of the researchers only reaching small and idiosyncratic networks, the likelihood of those who are socially isolated being excluded, and those who are more vocal or prominent in the community being over-represented, and the potential for findings to be strongly affected by the quality of the researcher (Griffiths et al., 1993). The Drug Transitions study sought to overcome these issues by recruiting PAIs from a range of backgrounds, all of who would have access to a wide selection of networks, and by developing a structured questionnaire to guide the interview.

The questionnaire collected basic demographic data, current patterns of drug use, history of drug-using behaviour, transitions in route of administration, social factors associated with transitions or non-transitions in route of administration, injection and sharing practice, details on living situation, and current and past sexual behaviour. PAIs were trained in the use of the interview schedule and, to ensure

data quality, all interviews were recorded and checked by the research team (with the caveat that recorders could be turned off during the interview if participants requested).

Ethics

Ethical approval was gained from the Joint Ethics Committee of the Institute of Psychiatry (prior to it becoming part of King's College London) and the Bethlem & Maudsley NHS Trust (the former name of SLAM).

2.4.2 Measures

Initial Route of Heroin Administration (IAR)

Participants were asked "Thinking back to the first time you used heroin:

- did you inject it?
- did you chase it?
- other

Of the 408 participants, 231 reported chasing as their IAR and 106 reported injecting. 'Other' was reported by 63 participants, of whom 58 reported snorting heroin, 3 reported smoking heroin and 2 reported oral administration. The IARs of injecting, chasing and snorting were included in the analyses, with smoking and oral excluded due to very low group numbers.

Speed of Transition Variable

Speed of Transition to Daily Use

Participants were asked "how long was it from your first use of heroin until it became something you used every day or most days?" Responses were provided in days, months or years, and were then recoded into the categories 1 to 3 weeks (i.e. up to one month), 1 month to 11 months, 1 to 2 years and 2 or more years. Individuals who had reported not progressing to daily heroin use at time of interview (N=34) were not included in the analysis.

Covariates

Gender

Data on gender were obtained through interviewer report. The sample was predominately male (61.5%, N=246).

Ethnicity

Participants reported being White, African/Caribbean, Asian, or Mixed. Due to low numbers of Black and Minority Ethnic participants this was coded as a binary White/Non-White variable. The sample was predominately white (90.1%, N=347).

Daily Other Drug Use Prior to Heroin Initiation

Data were obtained through self-report to the item "when you used heroin for the first time, were you using any other drugs regularly (that is every day or most days)?" The majority of the sample reported regular use of other drugs at heroin initiation (68.7%, N=274). Reported drugs included amphetamine (N=85), cannabis (N = 232), cocaine (N = 59) and ecstasy (N = 9).

Year of Initiation of Heroin Use

This was calculated from recorded year of interview, participant age at interview and age at initiation of heroin use. Year of initiation ranged from 1954 – 1991 (mean = 1981, s.d. 6.4).

Current Treatment

Data were obtained through self-report of attending a drug clinic, a street agency, a needle exchange, or receiving treatment for a drug problem somewhere else at the time of interview. As a result of the sampling method (i.e. by design) just over half the sample (58.5%, N = 234) was receiving treatment at time of interview.

2.4.3 Missing Data

There were 13 participants with missing data on daily heroin use status. These participants were not significantly different to those who had reported their time to daily heroin use on gender (P=0.92, OR 1.06, 95% CI 0.34 - 3.31), ethnicity (P=0.21, OR not possible due to 0 cell counts), year of heroin onset (F=0.00, d.f=1, P=0.97), or IAR (chasing P = 0.87, OR = 0.90, 95% CI 0.26 - 3.15; snorting P = 0.52, OR = 46, 95% CI = 0.52 - 4.46). No participants were missing data on their IAR. Information on missing data for all independent and dependent variables and covariates is available in Appendix 2.

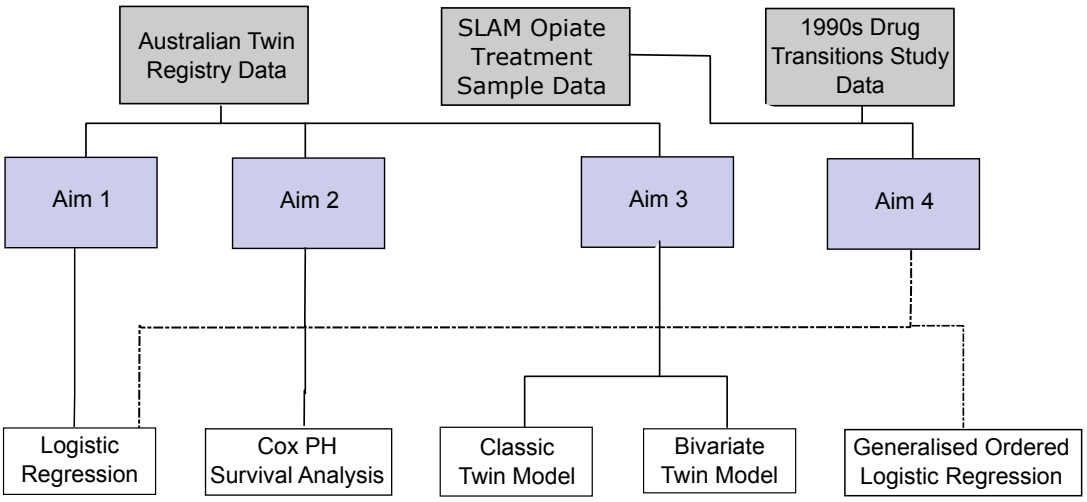
2.5 Analysis of Data

This section provides an overview of how analyses will address the aims of the thesis. See Figure 2.9 for an outline of analyses by aims addressed.

The results chapters (Chapter 3, 4, 5 and 6) will contain individual analysis plans outlining the specific aims of the analyses in relation to that chapter, how measures are used within each analysis, identified confounding variables, and any further specific details of the analysis.

The following statistical methods are employed throughout the thesis. The aim that the method is addressing in each chapter will be outlined, followed by an overview of the methodology, a description of the assumptions, and exposition of the complexities of the analysis as applied in this thesis.

Figure 2.9: Datasets Used for Each Aim, and the Analyses Applied to Address Aims



2.5.1 Data Preparation

Due to the size of the dataset, ATR data were initially cleaned and coded into the required variables using SAS software (SAS Institute Inc, 2002). Epidemiological analyses were conducted in Stata (StataCorp, 2009) and genetically informative analyses were conducted in R (R Core Team, 2013) using the package OpenMx (Boker et al., 2011). For the SLAM Opiate Treatment Sample and 1990s Drug Transitions Study data, all variables were coded and analysed using Stata (StataCorp, 2009).

2.5.2 Regression Models for Categorical Data

Aims Addressed

Regression analyses were used

1. in Chapter 3 to explore the association between early opportunity to use cannabis and the later outcomes of daily cannabis use, abuse/dependence and treatment-seeking;
2. in Chapter 3 to explore the extent to which the speed of transition to cannabis opportunity and from opportunity to dependence are influenced by individual, childhood, mental health and other drug use factors;
3. in Chapter 5 to explore the association between speed of transition from first to second use of cannabis and the later outcomes of daily cannabis use, abuse/dependence and treatment-seeking;
4. in Chapter 6 to explore the association between early transitions in heroin use and the later heroin use outcomes of heroin dependence severity, overdose, injecting behaviours and heavy heroin use, and the relationship between IAR and speed of transition.

Regression Outline

Regression analysis is used to determine the association between one or more factors and a single outcome (Vittinghoff et al., 2005), and can be used to establish the change in an outcome, or Dependent Variable (DV) for every one unit increase in a predictor, or Independent Variable (IV) (Vogt and Johnson, 2016). The size of this change is represented through the regression coefficient, which in analysis of dichotomous data can be calculated as an Odds Ratio (OR) through exponentiation of the regression coefficient. An OR of 1.0 indicates no relationship between the variables, and the difference from 1.0 in either direction provides an estimate of whether the relationship is positive or negative, and what the size of the effect is (Vogt and Johnson, 2016). Precision of the estimate can be assessed through 95% confidence intervals (95% CI).

Forms of Regression Used in the Thesis

Binary Logistic Regression

Logistic regression is appropriate for use when the DV is dichotomous, such as being a case or a non-case for cannabis dependence. The IV can be either binary or have three or more categories, and provides regression coefficients that compare each level of the independent variable against a set reference category. In this thesis, this reference category is the group with the least risk under the study hypothesis. When the IV has 3+ levels, differences between levels can be assessed through the use of post-hoc tests, such as Wald χ^2.

Ordered Logistic and Generalised Ordered Logistic Regression

When the DV has three or more levels and is an ordered variable, an ordered logistic model is appropriate (Williams, 2006). Under the proportional odds assumption the relationship with the IV must be the same for each level of the DV. This can be tested using likelihood ratio tests and the Brant test, with a significant result ($P \leq 0.05$) indicating assumptions are violated (Williams, 2006).

When the assumption is violated a generalised ordered logistic model can be used (Williams, 2006; Miller et al., 2015), which allows a different relationship between the DV and each level of the IV. This analysis produces multiple coefficients as the levels (J) of the DV are analysed equivalent to a series of binary logistic regressions where the categories (M) of the DV are combined. For example when M = 4, for J = 1 category 1 is contrasted with categories 2, 3, and 4; for J = 2 the contrast is between categories 1 and 2 versus 3 and 4; and for J = 3, it is categories 1, 2, and 3 versus category 4 (Williams, 2006).

2.5.3 Survival Analysis and Cox's Proportional Hazards Model

In order to identify factors that influence the speed of transition in stages of cannabis use, survival analysis was utilised for variables where continuous time data were available. Survival analysis uses regression analysis for time-to-event data to determine how many individuals in a sample will "survive" (not develop the outcome) past a certain time point (t), and allows for the inclusion of covariates to

explore how other factors affect the probability of "survival" (Fisher and Lin, 1999; Kleinbaum and Klein, 2012). This is represented through the hazard function (h(t)), which is the potential per time unit for an event to occur, given that the individual has survived up until t. Survival analysis uses time data h(t) as a rate rather than a probability or likelihood (Kleinbaum and Klein, 2012). The regression coefficient for survival analysis is the hazard ratio, which compares the hazard function for unexposed and exposed individuals in order to provide an estimate of the hazard for each group (Kirkwood and Sterne, 2003). This hazard represents an individual's increased rate of progression to the outcome.

The Proportional Hazards Assumption

A key assumption underlying survival analysis is the proportional hazards assumption: the hazard ratio does not vary with time. If this assumption is breached by covariates in the model, the relative risks of covariates will be incorrectly estimated, with increasing hazard ratios over time resulting in over-estimation and converging hazards resulting in an underestimation (Schemper, 1992).

There are a number of equally suitable options for assessment of the proportionality of the assumption (Bellera et al., 2010), and in this thesis, as in previous research (Waldron et al., 2014), analysis of Schoenfeld residuals has been selected. The residuals quantify the differences between what has been observed in the data and what would be expected under the model assumptions, and are calculated for every failure time (Bellera et al., 2010). A significant Pearsons Rho ($P \leq 0.05$) indicates breach of the proportional hazards assumption. When covariates do breach the assumption, the interaction with time has been included in the model in order to adjust for the breach of assumptions (Bellera et al., 2010; Schemper, 1992; Waldron et al., 2014). This results in an "extended" cox model.

Censored Data

One key feature of survival analysis is the use of censored data, whereby data that are missing after a certain time point are analysed without introducing bias through lost information (Kleinbaum and Klein, 2012). Censoring most commonly occurs when a subject does not experience the event before t, when a subject is lost to follow-up, or when a subject withdraws from the study. Given that the analyses in this thesis are retrospective, data are right censored: the outcome of interest has not occurred by t (time at interview). As there is still potential for the

outcome to occur after this time point, data are censored for analysis rather than being analysed as if the individual would never experience the outcome.

Person-Year data

Person-year data sets were constructed for the analyses, providing a separate row of participant data for each year so that there are as many data rows as there are time intervals at risk of the event occurring for each person (Jenkins, 2008). In order to create person-year data in Stata, the stsplit command was used once survival data had been created. This created a separate row of data for each year in which the participant was included within the analysis. In order to account for multiple participants experiencing failures events in the same year and having identical failure times, the Efron adjustment for survival ties was applied (Efron, 1977).

Time-Varying Covariates (TVC)

Where temporal data were available Time Varying Covariates (TVC) were created. In this thesis, TVC have been used to create a “step function” (Fisher and Lin, 1999); the hazard associated with the covariate will alter in the analysis at the point of covariate onset. If visualised, the hazard would "step" up or down (depending on the association with the outcome) at the point of onset. To achieve this, the Stata stsplit command was used once survival data had been created. This allows for individuals to be coded as 0 at the time-points before the reported onset of the behaviour of interest, and 1 at and forwards from the reported onset of the behaviour, and thus provides a more accurate representation of the individual hazards over time.

2.5.4 Additional Regression Considerations

Huber-White Adjustment for Clustered Data

The non-independence of observations from members of a twin pair or from participants recruited from within the same treatment clinic, can violate the assumptions of regression analyses. Therefore, the Huber-White analysis for clustered data was implemented in all regression and Cox PH analyses for data from the ATR (clustering by family) and the SLAM Opiate Treatment Sample (clustering by clinic). Clustering data were not available for the 1990s Drug

Transition Study. This method ensures robust standard errors (Maas and Hox, 2004).

Selection of Potential Confounding Covariates

The regression equation allows for multivariate modelling, whereby additional covariates are added to the regression model to allow the effect of the independent variable on the dependent variable to be assessed after accounting for the influence of these covariates. Doing so statistically adjusts the variables in the model for their differences in distribution and their relationship to the other variables in the model, providing estimates that are adjusted for the other variables (Hosmer et al., 2013). This method allows the model to account for confounding variables. These are covariates which are associated with both the independent and dependent variables, can therefore account for part of the association between the variables of interest, and may lead to incorrect estimation of the true association if not handled correctly (Hennekens and Buring, 1987).

For the logistic and generalised ordered logistic regression analyses of the ATR data (Chapter 3 and 5) potential covariates (described above in ATR measures, section 2.2.2) were selected based upon the literature (see Chapter 1) and their availability within the data set. In order to determine appropriate adjustment for each analysis model, all study covariates were entered into multivariable regression models with the variables age of opportunity to use cannabis, speed of transition from initiation to subsequent cannabis use, daily cannabis use, cannabis abuse and/or dependence, and cannabis treatment-seeking. This method is preferable to bivariable analysis (testing associations between covariates and IV/DV singularly), through which significant associations can be masked (Sun et al., 1996). Using multivariable regression allows associations to be adjusted for the effect of the other covariates, and the results were used to select confounding variables for analysis. Covariates that were identified as significantly associated with the model IV and DV were included to adjust for the potential confounding variable. The associations between the model IV and the covariates are reported in the relevant chapter (Chapters 3 and 5), and the associations between DV and the covariates are reported in Appendix 3. No ATR data analyses were adjusted for age at interview as the twin cohort was a birth cohort (born 1972 - 1979), resulting in little variation in age.

For the data from the SLAM Opiate Treatment Sample and the 1990s Drug Transitions Sample, a much smaller range of covariates was available. Analyses were adjusted for demographic factors, year of heroin onset to account for any cohort effects, and where possible, treatment status and regular other drug use preceding heroin initiation.

2.5.5 Twin Modelling

Twin modelling was used in Chapters 4 and 5 to address the aim of

1. examining the extent to which the speed of early stage transitions in trajectories of cannabis use were influenced by additive genetic, shared and non-shared environmental influences
2. exploring the extent to which genetic influences transitions are unique to the phenotype, and the extent to which they are correlated with cannabis dependence.

The Underlying Theory Of Twin Modelling

Underlying twin modelling is the concept of heredity – traits from parents are passed down to offspring through genes. Between any two individuals 1 in 1000 genes will have a polymorphism that differs, and these alternative forms of genes are referred to as alleles (Neale and Cardon, 1992). One allele can be dominant over another, which can determine which traits are expressed. Differences in genetic variants result in variation in physical traits such as eye colour, disease propensity, and individual behaviour. A person's complete collection of genes is referred to as the genotype, and observable characteristic or behaviour is termed the phenotype (Neale and Cardon, 1992).

Research has indicated that there are not singles genes that influence mental health and behavioural phenotypes. Rather, influences from a number of genes combine to produce outcomes such as drug dependence (Plomin et al., 2013). Consequently it is important to consider the combined effect of genes on a phenotype. This can be achieved by looking at the "variance" of a phenotype: the measure of individual differences in the population (Plomin et al., 2013).

The proportion of the phenotypic variance that can be accounted for by genetic differences amongst individuals is known as heritability (Plomin et al., 2013).

Not all variance in a population is attributable to genetic effects. There is a strong role played by the environment in mental health and behavioural outcomes. For example, it is impossible for genes to contribute to the development of dependence if an individual does not have access to the drug. This drug access can only come from the environment (and is only one example of many environmental influences). Environmental variance is partitioned into genetic and environmental sources through twin modelling.

The Role of Monozygotic (MZ) and Dizygotic (DZ) Twin Pairs

If phenotypic variance can result from both genetic and environmental differences, how can these be disentangled? Approaches using twins reared together can be used to determine the heritability of, and environmental contribution to, a phenotype or trait. Identical – or monozygotic (MZ) – twins pairs share 100% of their genetic material. Conversely fraternal – or dizygotic (DZ) – twin pairs share only 50% of the same genetic material. This means that they are no more alike, genetically, than full siblings. However, unlike siblings, DZ twins will grow up in the same environment. Using this knowledge we can calculate the extent to which the variance in a phenotype is due to genetic effects, and the extent to which it is due to environmental effects (Plomin et al., 2013). An MZ twin concordance that is greater than DZ concordance indicates that there is evidence for heritability, whereas MZ twin concordance that is less than 1 provides evidence for the effect of the environment on the trait. The different components of this variance, and further details on twin concordance, are outlined below.

Variance Components

Genetic Effects (A or D)

Genetic influences are indicated when within-pair concordance is greater in MZ twin pairs than in DZ twin pairs. Twin modelling can estimate the additive genetic effect (A), which is the sum of the effects of multiple genes on the phenotype. Increased within-pair similarity in MZ twins relative to DZ twin pairs is assumed to indicate that a greater proportion of the variability in the trait can be attributed

to genetic effects, and if the MZ correlation is twice the DZ correlation then all twin-pair similarity can be attributed to A (Verweij et al., 2012).
It is also possible to estimate the effect of interactions between alleles within genes (dominant genetic effects, D) (Neale and Cardon, 1992). If the MZ correlation is more than twice the DZ correlation, non-additive genetic factors may be involved. This component can also include epistatic variance – the effect of interactions between genes (Verweij et al., 2012). In studies of twins reared together it is not possible to estimate shared environmental influences (see below) at the same time as D, as the two variance components are confounded.

Shared Environment Effects (C)

The shared environment (C) refers to the non-genetic influences that lead to similarity amongst twin pairs (Plomin et al., 2013). These can include characteristics of parents such as socioeconomic status, and the neighbourhood a child is brought up in. If the MZ correlation is greater than the DZ correlation, but not twice the DZ correlation, there is evidence of some effect of C on the variance.

Unique Environment Effects (E)

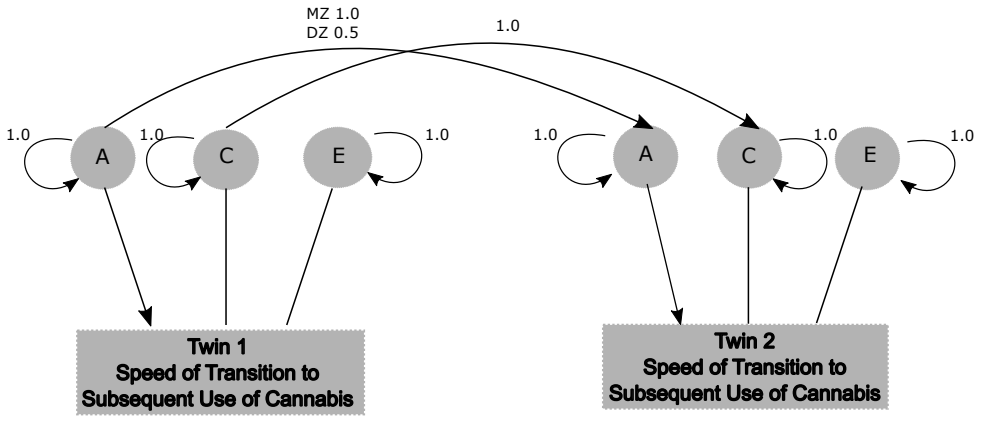
The unique environment (E) refers to non-genetic effects that are independent amongst twin pairs (Plomin et al., 2013). This can include friendship groups, and differences in treatment by family members. The extent to which the MZ twin correlation is less than 1.0 indicates the magnitude of non-shared environmental influences. As MZ twins share the same A and C effects, differences within these pairs (and also measurement error) are attributed to E (Plomin et al., 2013).

Obtaining Variance Estimates Through Twin Modelling

Structural equation modelling of twin data is used to obtain precise estimates of A, D, C and E and allows for the inclusion of effects such as sex and age, the comparison of models, and the generation of confidence intervals around estimates (Verweij et al., 2012). Using the R analysis package OpenMx (Boker et al., 2011) allows the parameters of the variance components to be estimated by use of matrix algebra. Matrices are created containing the expected variance (the differences in the phenotype within the whole population) and the expected covariance (the relationship between two characteristics studied; in twin modelling, the covariance of the observations within twin pairs) (Plomin et al., 2013). The

expected covariance is set to 1.0 for MZ twins and 0.5 for DZ twins, representing the known genetic correlation within these pairs. An outline of this, showing the variance components utilised in thesis analyses, can be seen in Figure 2.10. These expected variance-covariance matrices are compared to the observed variance and covariance. Maximum likelihood estimation, based on the assumption that observed variables have a multinormal distribution (Neale and Cardon, 1992), maximises the extent to which the model fits the data available by testing multiple possible model solutions. The model solution that provides the optimum fit to the data is selected (Verweij et al., 2012).

Figure 2.10: Path Diagram of Classic Twin Model, Indicating the Covariance of A and C Between MZ and DZ Twin Pairs



Tests of Model Fit

The methods used to assess model fit in this thesis are the Akaike Information Criterion (AIC) and the -2 log-likelihood ratio (-2LL). The AIC is a measure of the fit of a model that can be used to aid model selection, and utilises assessment of goodness of fit whilst also applying penalties for the number of parameters in a model. As such, this measure favours the most parsimonious models. The distribution of -2LL approximates a χ^2 distribution with degrees of freedom equal to model parameters. When assessing a model, a lower -2LL indicates a better model fit (Neale and Cardon, 1992).

Nested Models

In order to identify the most parsimonious model, models can be "nested". Once a model has been specified, nested models with different restrictions (but the same underlying specification) are compared against it. The -2LL of a model is especially useful when assessing nested models as it can be used to test differences in the fit of more parsimonious models compared to the saturated or ACE model ($P \leq 0.05$ indicating that there are significant differences in the fit of the two models). An increase in -2LL and the accompanying degrees of freedom between nested models would suggest that model fit had degraded rather than improved. In this thesis the -2LL is used to assess the difference in model fit between a saturated model and a model with equated parameters or thresholds, between an ACE model in which all the parameters are freely estimated and a more parsimonious version of the same model with certain parameters equated or fixed to a certain value, and between a model with different parameter estimates for males and females and one in which these parameters are equated across sexes.

The Liability Threshold Model For Ordinal Twin Data Analysis

Ordinal data analysis is required when the data used are not continuous, as is the case for the analyses in this thesis. In this circumstance a liability threshold model is used. The threshold model for ordinal data assumes that the liability to an outcome has an underlying normal distribution , but that the outcome is considered to occur once a certain point is reached (Plomin et al., 2013).

The thresholds in the model are determined through the use of z-values, which ensure that the area under the standard normal distribution curve between two thresholds reflects the prevalence of that category within the population (Posthuma et al., 2003). The proportion in each sample is calculated cumulatively: the N in the first category is estimated as a percentage (e.g. 40% of sample), setting the first threshold. The next threshold will be the proportion in that group added to the proportion of the first. Z scores are calculated from these proportions and used to set the thresholds for analysis.

These thresholds are then used for the expected variance-covariance matrices in the twin model. Correlations between ordinal variables, or between one ordinal and one continuous variable, are calculated as tetrachoric (2 variables) or

polychoric (3+ variables) correlations (Neale and Cardon, 1992). The expected and actual variance-covariance matrices are then compared and parameters estimated, as outlined above. One implication for the interpretation of threshold model results is that power is reduced when compared to the analysis of continuous data (Neale et al., 1994). Nonetheless, this methodology is a viable and commonly used approach for categorical phenotypes.

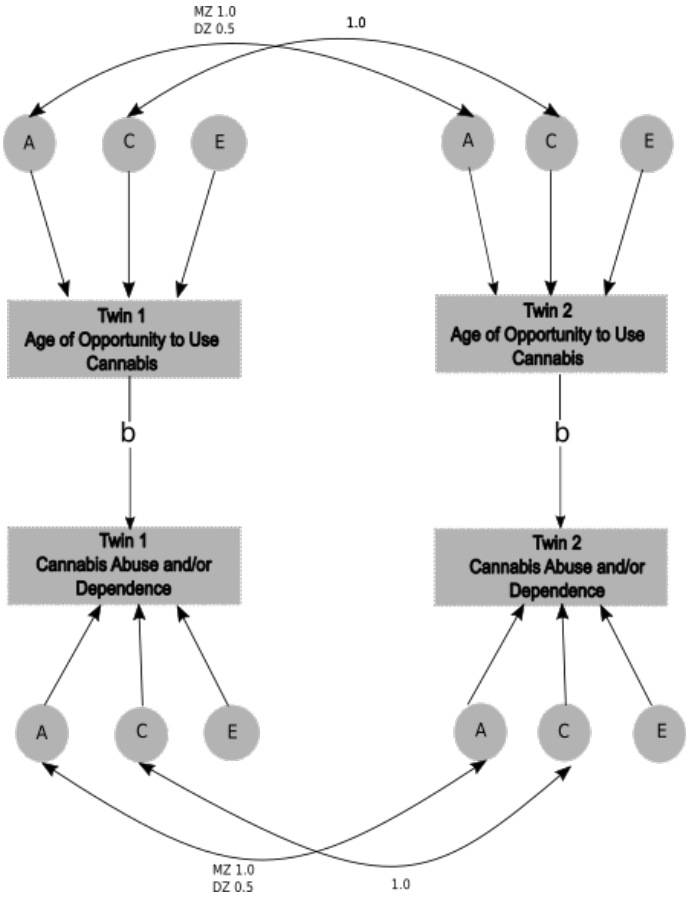
Multivariate Twin Modelling

Multivariate analysis extends twin modelling to allow the study of multiple traits, and to identify whether the traits share the same underlying causes (Plomin et al., 2013). By including two or more traits, the covariance across traits and between twins can be studied. For example, hypotheses can be tested relating to the correlation between drug use in one twin and problematic drug use in their co-twin. The correlation between the traits can be partitioned into the same A, C and E variance components as in univariate analysis. The multivariate analyses utilised in this thesis are outlined below.

Causal Contingent Common Pathway Model

Twin modelling can be used to test whether the influences underlying two related phenotypes are correlated. When an outcome cannot occur unless an earlier stage has been reached – for example, an individual cannot report cannabis dependence if they have not had the opportunity to use cannabis – a Causal Common Contingent Pathway (CCC) extension to classic twin modelling can be applied. See Figure 2.11 for an outline of the model. This model tests whether liabilities to the study outcomes are consistent with a single liability distribution (all the same underlying factors), or a multiple liability distribution (underlying factors independent or correlated) (Maes et al., 2004b). Conceptualising the phenotypes as related through a process of stages (Neale et al., 2006b), an estimation of the magnitude of the relationship between the two phenotypes studied is provided through a beta pathway (Fowler et al., 2007). If the beta estimate is 1 this suggests that the two phenotypes completely overlap – a single liability distribution (Do et al., 2015; Fowler et al., 2007). However, whilst this model can identify whether liabilities are correlated, it cannot determine the extent to which this correlation is attributable to genetic or environmental influences.

Figure 2.11: Causal Contingent Common Pathway Model Path Diagram

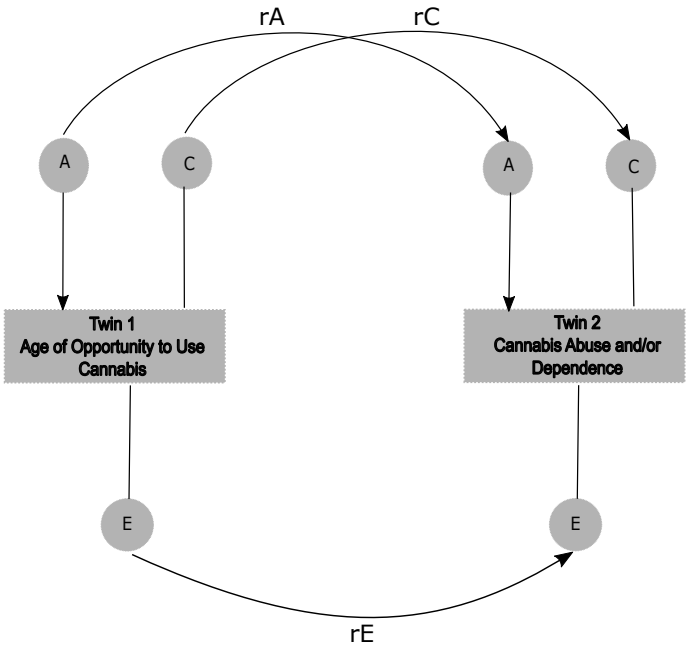


Two-Stage Bivariate Twin Model

Twin models can be extended to include more than one phenotype, and determine not only the variance components for those two or more phenotypes, but also the extent to which genetic and environmental influences on the phenotypes are correlated (Neale and Cardon, 1992). However, there are issues with this methodology for phenotypes that require an environmental exposure for them to occur; phenotypes such as drug dependence. Structural missing data (for example, individuals who have not had an opportunity to use cannabis are unknown/missing on liability to dependence) would preclude standard bivariate analysis of the relationship between two traits. It is not feasible to simply remove those who are not exposed from the analysis, as genetic and environmental factors that determine variation in dependence may also determine risk of exposure, and

excluding those unexposed from the analysis will discard genetic information and result in bias. Alternately, if genetic and environmental influences on risk of exposure are uncorrelated with influences on dependence, including non-users as non-dependent individuals would confound two traits that have different modes of inheritance (Heath et al., 2002).

Figure 2.12: Constrained Correlations Two-Stage Twin Model Path Diagram



Two-stage twin modelling has been developed to overcome this issue. As with a CCC model, the two-stage twin model conceptualises the phenotypes of interest as staged, but the analyses in this model allow estimation of the overlap in genetic and environmental influences between two phenotypes that have correlated liabilities. Developed by Heath et al. (2002), this model is used to estimate the correlation between A, C and E in situations where early-stage phenotypes, such as cannabis use opportunity, are *necessary* for the expression of later behaviours, such as the development of dependence. Operationalising the first stage phenotype as a multiple category trait with several ordered categories, rather than a binary trait, overcomes this issue. By including those who did not reach the first stage of the process (for example, those who never had the opportunity to use cannabis) the model maximises the genetically informative information available and avoids bias that can result from inappropriate classification on non-exposed individuals.

Once the data are coded as a multiple category trait, a standard Constrained Correlations model can be applied to the data. By utilising the cross-covariance within twin pairs the A, C and E parameters for each phenotype are estimated, and the phenotypic correlation partitioned into the genetic and environmental contributions to the correlation. See Figure 2.12 for outline of model.

Modelling Potential Sex Differences

Twin models can be modified to test for differences in the magnitude of genetic and environmental effects between sexes (scalar sex differences), and differences in the genetic and environmental factors influencing the phenotype between sexes (non-scalar sex differences) in the genetic and environmental parameters and correlations for two phenotypes (Neale et al., 2006c). Scalar sex differences are tested by examining whether constraining the estimates of A, C and E to be equal in males and females leads to a significant deterioration in model fit. Non-scalar sex differences are tested by constraining the A correlation to be 0.5 and the C correlation to be 1 in opposite sex DZ twin pairs. Given the genetic relatedness of DZ twin pairs the expectation is that the correlation between pairs will be 0.5. If the opposite sex twin pair correlation is below this it suggests that different genetic factors are involved for each sex. Similarly, the shared environmental correlation between DZ twins is expected to be 1.0, so a correlation below this for opposite sex twin pairs indicates that the shared environmental factors influencing the phenotype are not the same across sexes.

The Equal Environments Assumption (EEA)

The Equal Environments Assumption (EEA) is one of the key debates in twin research, and is commonly levelled as a critique of the methodology. The assumption states that, despite differences in genetic similarity, environmental influences will be equally correlated in both monozygotic and dizygotic twins. This is important as estimates of heritability will be inflated if there is greater similarity in MZ environments, as the heritability estimate is based on the principle that a greater correlation with an outcome in MZ twins compared to DZ twins results will

be due to the increased genetic similarity of MZ twins (Kendler et al., 1993).

A number of different methods have been used to test the EEA, including testing if physical similarity correlates with trait similarity (on the basis that similarity of treatment stems from physical similarity) (Plomin et al., 1976), observing in childhood whether the behaviour of others towards a twin is self-initiated or occurs in response to the twin's behaviour (Lytton, 1977), exploring correlations of childhood environment similarity and later adult behaviour and personality similarity (Heath et al., 1989), and studying twins who were misinformed of their zygosity to determine the influence of social expectations of similarity (Kendler et al., 1993). These studies suggest that twin resemblance is not influenced by physical similarity, excess resemblance in parental behaviour towards MZ than DZ twins stems from the behaviour of the twin rather than being initiated by the parent, consistent relationships have not been observed between childhood environment similarity and later patterns of personality and behaviour, and perceived zygosity does not influence twin resemblance for psychiatric disorders (Kendler et al., 1993).

The critique of twin modelling is that differences in environmental similarity between MZ and DZ twins will inflate estimates of heritability. In order to address this, research has investigated the relationship between environmental similarity and a number of psychiatric traits (Kendler and Gardner, 1998). In a cohort of female twins, measures of similarity were assessed through questionnaire and then grouped based on factor loadings into "childhood treatment", "co-socialization" and "similitude" (the degree to which twins viewed themselves to be alike). The study found that MZ twins scored significantly higher for co-socialization and similitude than DZ twins, but did not observe differences in childhood treatment. The study went on to explore the effect of this finding on correlations for a range of psychiatric outcomes (including alcohol and tobacco dependence). Associations between similarity of environmental experiences and later psychiatric outcomes were not found, with the exception of associations between co-socialization and smoking initiation, and co-socialization and broadly defined bulimia. This indicates that, even in situations where there is inequality in environmental similarity, this does not impact on association with later psychiatric outcomes.

Testing the EEA

As the analyses in this thesis are based on retrospective self-report, many of the methods mentioned above to test the EEA are not feasible. However, questions

were available on the perceived similarity of childhood environments (see Section 2.2.2). It was therefore possible to compute heritability estimates for twins reporting similar or dissimilar childhood environments. The twin sample was split into two, based on whether or not twins were concordant on reported childhood environment similarity, and ACE models were run on these two separate data groups. If violations of the EEA inflate estimates of heritability, it would be hypothesised that heritability estimates would be higher for those raised in similar environments. The method used to test the EEA in this thesis was based on that used to test the assumption in a similar sample of Australian twins (Lynskey et al., 2002).

Testing the EEA for the outcome of cannabis abuse and/or dependence, it was found that estimates were similar across twins with equal and unequal environments. Amongst those reporting equal environments parameter estimates were A = 0.64 (95% CI 0.22 – 0.89), C = 0.16 (95% CI 0.0 – 0.50) and E = 0.20 (95% CI 0.11 – 0.35). For those reporting unequal environments parameter estimates were A =0.76 (95% CI 0.40 – 1.00), C = 0.0 (95% CI 0.0 – 0.28) and E = 0.24 (95% CI 0.12 – 0.42). All parameter estimates fall within the 95% CI for either group, indicating that equality of environment does not affect estimates for cannabis abuse and/or dependence in this sample.

Model Adjustment for Covariates

Twin modelling analyses can be adjusted for variables such as sex, in order to prevent overestimation of the intraclass twin correlation. In order to do so in OpenMX, a separate matrix is created for the sex definition variables, and the estimated parameters are obtained by regressing the twin data on the sex variables (McGue and Bouchard, 1984).

CHAPTER 3

Begin at the Beginning: Age of Opportunity to Use Cannabis and Later Cannabis Use, Abuse and Dependence

3.1 Introduction

Opportunity to use a drug is the first stage of drug involvement, and a necessary first stage for the development of SUDs. Importantly, while opportunity is necessary for use to occur, use of the drug is not necessary for a valid opportunity to have happened. Opportunity has been defined in the literature as being offered the drug, or being present at a time when others were using and it would have been possible for the individual to use the drug if they had wanted to (Storr et al., 2011).

A limited number of factors have been studied in relation to the speed of transition to cannabis use opportunity (with earlier age at first opportunity representing a faster transition, given that there are no preceding stages of drug involvement). To date, few factors have been explored in relation to this. Disruptive behaviour early in school in males and better reading scores in females was found to be associated with younger age of opportunity to use cannabis (Storr et al., 2011). The findings on early disruptive behaviour are consistent with large body of literature, but the association with better reading scores may appear

counterintuitive. Nonetheless, it is consistent with recent research demonstrating that adolescents with higher academic ability at age 11 are more likely to use cannabis in high school (Williams and Hagger-Johnson, 2017). However, the Storr et al. (2011) study was limited by the lack of information on later childhood and adolescent factors. Lifetime regular tobacco use (Agrawal et al., 2013) has been found to be associated with earlier cannabis use opportunity. There is scope to understand this transition better by testing associations with a wider range of factors. Doing so will also allow comparison with factors that are associated with progression to dependence, and the identification of whether factors associated with problematic use begin to exert an influence on drug use behaviour at even this early stage.

Additionally, the relationship between age of opportunity to use cannabis and later cannabis use outcomes has not been explored in detail, whereas early initiation of cannabis use has been shown to be related to later cannabis abuse/dependence (Swift et al., 2008; Grant and Dawson, 1998; Fergusson and Horwood, 1997). Consequently little can currently be said about the relationship between age of opportunity to use cannabis and the relationship to cannabis abuse or dependence.

In this chapter I will identify factors associated with faster transition to opportunity to use cannabis, and whether these factors overlap with those that are associated with progression to cannabis dependence. I will also explore whether there is an association between age of opportunity to use cannabis and later cannabis use outcomes.

3.2 Aims and Hypotheses

Aim 1

Determine socio-demographic, childhood, mental health, peer and drug use factors that are associated with faster transitions to cannabis use opportunity, and from cannabis use opportunity to cannabis dependence.

Hypothesis 1

Multiple socio-demographic, childhood, mental health, peer and licit drug factors will be associated with more rapid transition from cannabis use opportunity, and from cannabis use opportunity to cannabis dependence.

Aim 2
Establish whether there is consistency in the socio-demographic, childhood, mental health, peer and drug use factors that are associated with faster transition to opportunity to use cannabis and those associated with more rapid progression from opportunity to dependence.
Hypothesis 2
The associated socio-demographic, childhood, mental health, peer and drug use factors will differ across the stages of cannabis use transitions.

Aim 3
Explore the relationship between age of opportunity to use cannabis and later cannabis daily use, abuse and/or dependence, and treatment seeking when accounting for the influence of individual, childhood, mental health, peer and licit drug factors that may confound the association
Hypothesis 3
Individuals who have faster transition to opportunity to use cannabis, represented through earlier age of opportunity, will have an increased likelihood of cannabis daily use, abuse and/or dependence, and treatment seeking later in life.
Hypothesis 4
Associations between speed of transitions from birth to opportunity to use cannabis and lifetime risks of cannabis daily use, abuse and/or dependence, and treatment seeking will persist after controlling for identified individual, childhood, mental health, peer and drug use factors identified as exerting a potentially confounding influence on the association.

See Figure 3.1 for an outline of how the hypotheses of this chapter are addressed through two different analyses.

3.3 Sample

The sample for all analyses in this chapter was drawn from the Australian Twin Registry (ATR). The 3798 participants who provided information on whether or not they had ever had the opportunity to use cannabis form the analysis sample in this chapter (see Figure 3.2).

Figure 3.1: Dataset and analyses used to address the aims of this chapter

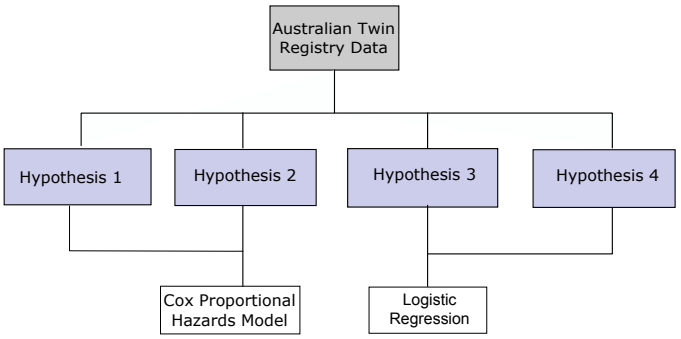
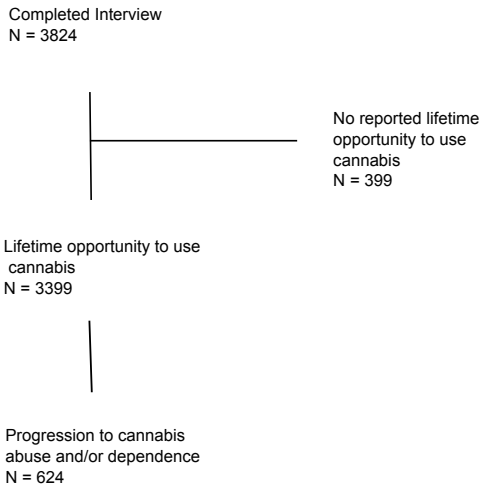


Figure 3.2: Deriving the analysis sample from the complete twin cohort



3.4 Factors Associated With Faster Transitions To Cannabis Use Opportunity, And From Cannabis Use Opportunity To Cannabis Dependence

The following methods were applied in order to study the factors associated with faster transitions to cannabis use opportunity and, in order to allow for study of the consistency of these influences across transitions, from opportunity to use cannabis to the development of cannabis dependence.

3.4.1 Methods

Exposure and Failure Variables

Age of opportunity to use cannabis

A continuous variable of age of opportunity was used in the analysis. The underlying timescale was in years, and data were normally distributed (see Chapter 2 for histogram).

Cannabis Dependence

A continuous measure of age at onset of cannabis dependence was used in the analyses. The underlying timescale was in years, and data were normally distributed (see Chapter 2 for histogram).

Analysis Plan

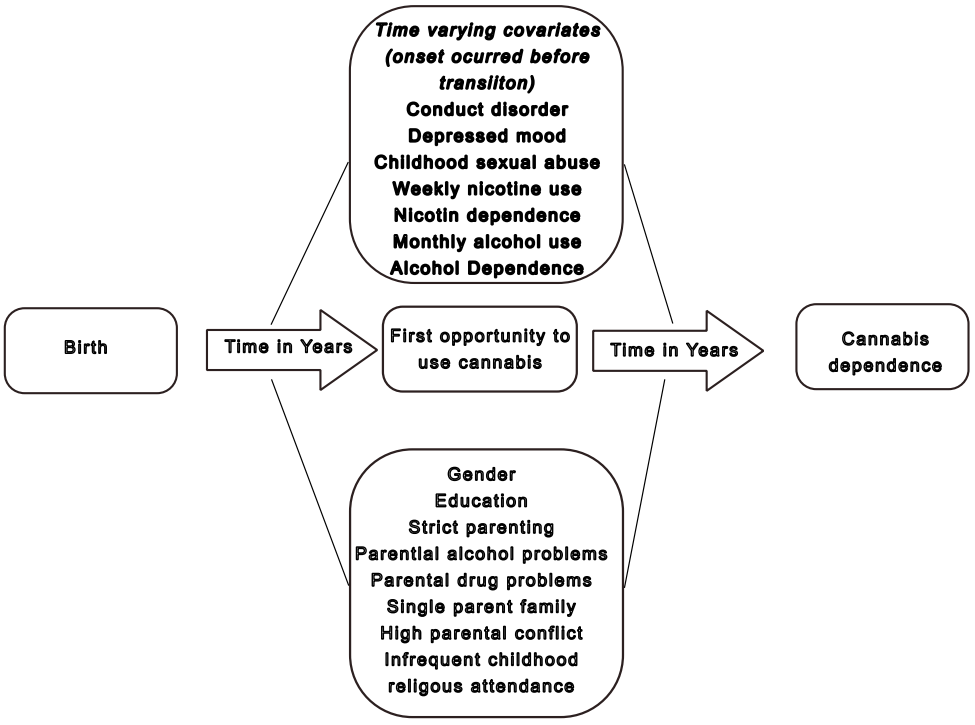
Survival analysis was used to identify factors that were associated with transition from 1) birth to opportunity to use cannabis 2) opportunity to use cannabis to cannabis dependence. A separate Cox Proportional Hazards (Cox PH) model was fitted to the data for each transition. See Figure 3.3 for an outline of the models. Analyses were conducted in Stata statistical software version 11 (StataCorp, 2009).

This method uses regression analysis for time-to-event data to determine how many in a sample will 'survive' past a certain time point, and allows for the inclusion of covariates to explore how other factors affect the probability of survival (Fisher and Lin, 1999; Kleinbaum and Klein, 2012) Speed of transition was assessed as time in years, providing a continuous time-to-event variable suitable for this analysis. The regression coefficient for survival analysis is the hazard ratio, which compares the hazard function for unexposed and exposed individuals in order to provide an estimate of the increase or decrease in speed of transition to outcome for each group (Kirkwood and Sterne, 2003). To correct for the non-independence of observations (due to family clustering), Huber-White adjustment for clustered data was implemented to provide robust standard errors.

Data Preparation and Assumption Testing for Cox PH Models

Person-year datasets were constructed to provide a separate row of participant data for each year from birth for model 1, and for each year from age of opportunity for model 2. In order to account for participants experiencing onset of events in the same year, the Efron adjustment for survival ties was applied (Efron, 1977). The

Figure 3.3: Cox PH models for factors affecting speed of transition from 1) birth to opportunity to use cannabis and 2) opportunity to use cannabis to cannabis dependence



assumption of proportional hazards was assessed through tests of Schoenfeld residuals and modelling of the interaction of covariates with time in the analysis ($P \leq 0.05$).

Censoring of Data

Participants were right-censored at age of interview. Mean age of first cannabis use opportunity was 17.6 years (s.d. 3.2) and the mean age of cannabis dependence 21.4 years (s.d. 4.1). The mean survival time for the participants in the cannabis use opportunity model was 19.1 years (s.d. 5.1) (age at opportunity, or for those who did not report opportunity, age at interview). This figure is higher than the mean opportunity age as a result of right censoring; individuals who have not experienced opportunity by the point of interview are also included in the survival model, with their age at time of interview in place of age of opportunity. The mean survival time for participants in the cannabis dependence model was 13.4 years (s.d. 4.9) (time from opportunity to dependence, or for those who did not develop dependence, time from opportunity to age at interview). This figure is higher than may be expected from the mean dependence age, again as a result of right censoring; individuals

who have not developed dependence by the point of interview are also included in the survival time, with their time from opportunity to age at interview in place of time to dependence.

Cox PH model 1 - Identifying factors associated with speed of transition to opportunity to use cannabis

This model identifies factors associated with hazard of the opportunity to use cannabis. Survival data (time in years, starting from birth) were used for analysis of 3798 participants who had provided information on whether or not they had experienced opportunity to use cannabis. Failure event was opportunity to use cannabis, and 3398 failure events were observed. Due to missing covariate data, 3763 participants were included in the final model (3367 failure events).

Cox PH model 2 - Identify factors associated with speed of transition from opportunity to use cannabis to cannabis dependence

This model identifies factors associated with hazard of the development of dependence following the opportunity to use cannabis. Survival data (time in years, starting from age of first opportunity to use cannabis) were used for analysis of 2593 participants who had reported their age of opportunity to use cannabis and who had also reported lifetime cannabis use. The failure event was cannabis dependence, and 371 failures were observed. Due to missing covariate data, 2565 participants were included in the final model (363 failure events).

Fixed and time-varying covariates

From the full list of potential covariates selected from the literature (see Chapter 2, Section 2.2.2) Time Varying Covariates (TVC) were produced for those for which age of onset data were available. These were CD, depressive episode, CSA, monthly alcohol use, alcohol dependence, weekly tobacco use, tobacco dependence, other drug use, and other drug dependence. These variables were coded as present for each year after their respective age of onset, and were only included in the model if onset had occurred prior to the age of cannabis use opportunity for model one, or prior to the age of dependence for model two. For example, if a participant reported first opportunity to use cannabis at age 13, and reported tobacco onset at age 14, they would be coded as a non-case for tobacco use in model 1 as the TVC onset was later than the failure event.

Fixed covariates included in the model were gender, education, strict parenting, parental alcohol problems, parental drug problems, childhood religious attendance,

high parental conflict, and single parent family.

Level of completed education and school peer cannabis use were not included in the analyses as the temporality of these variables could not be determined (they may have reverse causality in the analysis). See Figure 3.3 for model outline, including fixed and TVC.

Observations removed from analysis

To minimise the likelihood that the effect of childhood covariates where the specified time periods were ages 6 – 13 (parental conflict, single parent family, strict parenting, frequent childhood religious attendance) may have occurred after the point of cannabis use opportunity, any individuals who reported use opportunity before the age of 6 were removed from model one. This resulted in the observations of one participant being removed from the model.

Those who had not reported lifetime cannabis use were removed from model 2 in order to avoid the inverse association that would exist between never-use of cannabis and cannabis dependence. Additionally, one participant was omitted from model 2 as their recorded age of dependence was earlier than recorded age of opportunity.

3.4.2 Results

Testing the Proportional Hazards Assumption

All covariates were tested for breach of the proportional hazards assumption, as outlined in the methods section (see Table 3.1). The following did not satisfy the proportional hazards assumption for the opportunity to use model and therefore the interaction term between the covariate and analysis time was modelled in the cannabis use opportunity analysis (Bellera et al., 2010): CD, parental drug problems, weekly tobacco use and monthly alcohol use. Similarly, for the cannabis dependence analysis the following variables had the interaction with analysis time modelled in the analysis: parental drug problems and alcohol dependence.

Table 3.1: Results of Proportional Hazards Assumption tests for all covariates, determined through tests of Schoenfeld Residuals and through modelling of the interaction with time in the analysis ((in the ATR sample, N=3798)

Covariates	Test of Schoenfeld Residuals	Test of Interaction With Time
	Pearsons rho	HR (95% CI)
	Birth to Opportunity Model	
Gender	0	1.00 (0.98 – 1.03)
Conduct Disorder	**-0.03***	**0.92*** (0.85 – 0.99)
Depressive Episode	0	1.00 (0.96 – 1.03)
High Parental Conflict	-0.01	1.00 (0.97 – 1.02)
Parental Alcohol Problems	0.01	1.01 (1.00 – 1.02)
Parental Drug Problems	**0.03***	**0.91*** (0.84 – 0.99)
Single Parent Family	0.01	0.99 (0.93 – 1.05)
Strict Parenting	0.01	1.01 (0.99 – 1.03)
Infrequent Childhood Religious Attendance	0.02	0.98 (0.96 – 1.01)
Childhood Sexual Abuse	0	1.00 (0.95 – 1.05)
Weekly Tobacco Use	**-0.06*****	**0.91***** (0.87 – 0.96)
Tobacco Dependence	0	1.06 (0.95 – 1.17)
Monthly Alcohol Use	**0.04***	**1.05*** (1.01 – 1.09)
Alcohol Dependence	0.02	1.05 (0.95 – 1.16)
Other Drug Use	0.02	1.02 (0.96 – 1.09)
	Opportunity to Dependence Model	
Gender	-0.02	1.02 (0.96 – 1.09)
Conduct Disorder	-0.02	0.99 (0.92 – 1.06)
Depressive Episode	-0.01	0.98 (0.93 – 1.05)
High Parental Conflict	0	1.00 (0.95 – 1.06)
Parental Alcohol Problems	0.03	1.01 (0.96 – 1.07)
Parental Drug Problems	**-0.12***	**0.89*** (0.80 – 1.00)
Single Parent Family	0.05	1.04 (0.93 – 1.15)
Strict Parenting	-0.01	1.00 (0.94 – 1.06)
Infrequent Childhood Religious Attendance	-0.04	1.03 (0.97 – 1.09)
Childhood Sexual Abuse	0.07	1.07 (0.98 – 1.16)
Weekly Tobacco Use	-0.02	0.98 (0.92 – 1.05)
Tobacco Dependence	-0.01	1.00 (0.93 – 1.06)
Monthly Alcohol Use	-0.08	0.93 (0.85 – 1.01)
Alcohol Dependence	**-0.16*****	**0.87***** (0.81 - .094)
Other Drug Use	0.09	1.07 (1.00 – 1.15)
Other Drug Dependence	-0.06	0.93 (0.82 – 1.06)

*P ≤0.05 **P≤0.01 ***P ≤0.001.

Factors Uniquely Associated with Opportunity to Use Cannabis

Results from the Cox proportional hazards model for transition to opportunity to use cannabis are presented in Table 3.2. High parental conflict (HR 1.09, 95% CI 1.00 - 1.18), parental alcohol problems (HR 1.19, 95% CI 1.08 - 1.30), CSA (HR 1.17, 95% CI 1.01 - 1.34) and infrequent childhood religious attendance (HR 1.35, 95% CI 1.26-1.45) were associated with increased hazard of earlier opportunity to use cannabis.

Factors Uniquely Associated with Progression to Cannabis Dependence

Results from the Cox proportional hazards model for transition from opportunity to use cannabis to dependence are presented in Table 3.2. Non-clinical depressive episode (HR 1.44, 95% CI 1.12 - 1.85), tobacco dependence (HR 1.36, 95% CI 1.04 - 1.78), alcohol dependence (HR 2.64, 95% CI 1.53 - 4.58), other drug use (HR 2.10, 95% CI 1.64 - 2.69) and other drug dependence (HR 2.75, 95% CI 1.70 - 4.43) were associated with increased hazard of faster transition cannabis dependence.

Factors Consistently Associated Across Transitions

Factors associated with increased hazard of both earlier cannabis use opportunity and faster progression to cannabis dependence were CD (opportunity HR 5.57, 95% CI 1.52-20.47; dependence HR 2.49, 95% CI 1.91 - 3.25), parental drug problems (opportunity HR 7.29, 95% CI 1.74 - 30.62; dependence HR 3.30, 95% CI 1.63 - 6.69), male gender (opportunity HR 1.43, 95% CI 1.33-1.54; dependence HR 2.33, 95% CI 1.83 - 2.96) and weekly tobacco use (opportunity HR 8.57, 95% CI 3.93 - 18.68; dependence HR 2.76, 95% CI 2.10 - 3.64) (see Table 3.2).

Results Summary 1

The analyses have identified a number of factors uniquely associated with progression to cannabis use opportunity and dependence, predominately representing proximal influences acting at the time of drug use. However, a number of consistent influences acting across stages have also been identified. These are predominately distal influences.

3.5 The Relationship between Age Of Opportunity To Use Cannabis And Later Cannabis Daily Use, Abuse and/or Dependence, And Treatment Seeking

The second part of this chapter focuses on identifying whether there is an association between age of opportunity to use cannabis and later cannabis abuse

Table 3.2: Hazard ratios (95%CI) from ATR Cox Regression Models: Factor Associated with Earlier Opportunity to Use Cannabis (N=3798), and for Progression from Opportunity to Use Cannabis to Cannabis Dependence (N=3797)

Covariate	Transition to Cannabis Use Opportunity N = 3763		Transition to Cannabis Dependence N = 3367	
	Unadjusted HR	Adjusted HR	Unadjusted HR	Adjusted HR
	(95% CI)	(95% CI)	(95% CI)	(95% CI)
Male gender	**1.45*** **	**1.43*** **	**2.00*** **	**2.33*** **
	(1.32 - 1.56)	(1.33 - 1.54)	(1.60 - 2.48)	(1.83 - 2.96)
Conduct disorder[1]	[2]**7.54*** **	[2]**5.57** **	**4.57*** **	**2.49*** **
	(2.39 – 23.76)	(1.52-20.47)	(3.63 – 5.75)	(1.91-3.25)
Depressive episode[1]	1.04	0.98	**1.95*** **	**1.44*** **
	(0.93 - 1.17)	(0.87-1.10)	(1.55 - 2.42)	(1.12-1.85)
High parental	**1.09***	**1.09***	1.16	1.02
conflict	(1.01 – 1.18)	(1.00-1.18)	(0.94 – 1.44)	(0.79-1.31)
Parental alcohol problems	**1.27*** **	**1.19*** **	**1.29** **	1.11
paroblems	(1.16 – 1.38)	(1.08-1.30)	(1.03 – 1.62)	(0.86-1.43)
Parental drug problems	[2]**8.26** **	[2]**7.29** **	[2]**4.14*** **	[2]**3.30*** **
problems	(2.12 – 32.15)	(1.74-30.62)	(2.07 – 8.27)	(1.63-6.69)
Single parent family	**1.30** **	1.13	**1.60***	1.19
family	(1.10 – 1.53)	(0.95-1.35)	(1.11 - 2.32)	(0.78-1.81)
Strict parenting	1.03	1.02	**1.32** **	1.11
	(0.96 - 1.10)	(0.95-1.09)	(1.07 – 1.62)	(0.88-1.39)
Infequent childhood	**1.35*** **	**1.35*** **	1.16	1.18
religious attendance	(1.25 - 1.46)	(1.26 - 1.45)	(0.93 - 1.44)	(0.94-1.48)
Childhood sexual	**1.25** **	**1.17***	**1.98*** **	1.35
abuse[1]	(1.08 – 1.42)	(1.01-1.34)	(1.49 – 2.64)	(0.95-1.92)
Weekly tobacco	[2]**10.17*** **	[2]**8.57*** **	**3.98*** **	**2.76*** **
use[1]	(5.00 – 20.71)	(3.93-18.68)	(3.12 – 5.07)	(2.10-3.64)
Tobacco	**1.82***	0.89	**2.77*** **	**1.36***
dependence[1]	(1.29 – 2.56)	(0.63-1.25)	(2.18 – 3.52)	(1.04-1.78)
Monthly alcohol	1.65	0.75	1.03	0.94
use[1]	(0.78 – 3.50)	(0.34-1.64)	(0.75 – 1.41)	(0.69-1.30)
Alcohol	**1.79*** **	1.26	**22.94*** **	**2.64*** **
dependence[1]	(1.29 – 2.48)	(0.89-1.78)	(1.69 – 5.12)	(1.53-4.58)
Other drug	**1.31** **	1.20	**2.76*** **	**2.10*** **
use[1]	(1.05 – 1.65)	(0.94-1.52)	(2.22 – 3.42)	(1.64-2.69)
Other drug	-	-	**5.70*** **	**2.75*** **
dependence[1]			(4.77 – 10.94)	(1.70-4.43)

*P≤0.05 **P≤0.01 ***P ≤0.001.
[1]Time Varying Covariate
[2]Interaction with analysis time included in the model
All models have applied the Huber-White adjustment for family clustering

and/or dependence. An association has previously been observed between age of initiation and later SUDs, but it is not known if this will extend to the stage before cannabis use has been initiated.

3.5.1 Methods

Independent and Dependent Variables

Age of Opportunity to Use Cannabis

Age of opportunity to use cannabis was coded into a categorical or dichotomous variable to provide meaningful estimates of effect size. The 10th, 25th, 50th percentiles of the distribution of the variable were used to determine transition speed groups (see Chapter 2 for distribution). This formed a four level categorical variable with the groups opportunity age 14 and under, opportunity age 15-16, opportunity age 17, and opportunity age 18 and over, which was used for all analyses in this chapter.

Cannabis Outcome Variables

The outcomes of daily cannabis use, cannabis abuse and/or dependence and cannabis treatment seeking were used in the logistic regression analyses (see Chapter 2 for derivation of these variables).

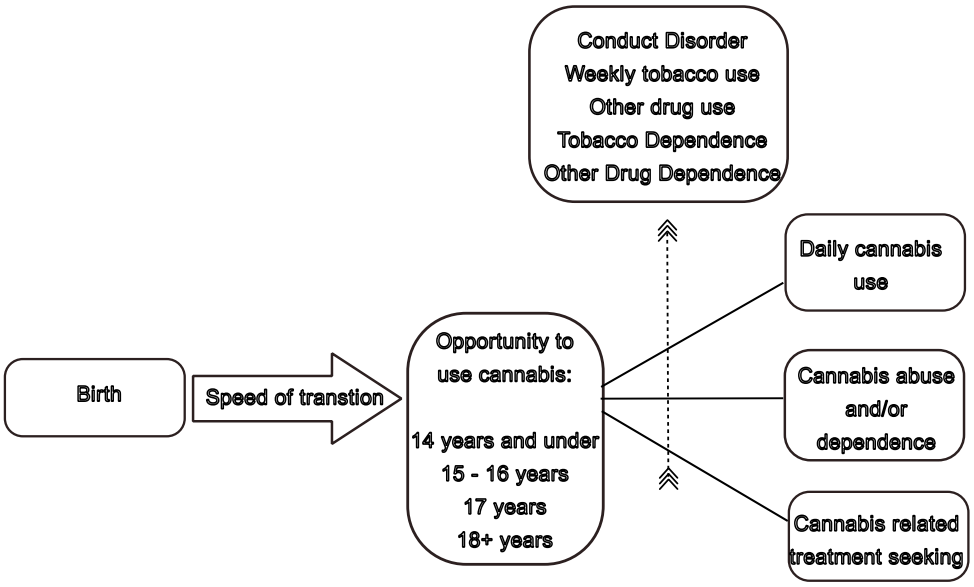
Analysis Plan

Logistic regression analysis was used to determine the association between the speed of transition to cannabis use opportunity and the following outcomes: (a) daily cannabis use; (b) cannabis abuse/dependence; (c) treatment-seeking for cannabis use problems. The reference category was the lowest-risk group, opportunity age 18 and over, based on the hypothesis that earlier opportunity to use is associated with greater likelihood of problematic cannabis outcomes. Figure 3.4 provides an outline of the analysis. To correct for the non-independence of observations Huber-White adjustment for clustered data was implemented to provide robust standard errors. Post hoc comparisons across the varying speeds of transition were conducted using Wald χ^2 tests. Analyses were conducted in Stata statistical software version 11 (StataCorp, 2009).

Identification of Covariates

From the full list of potential covariates selected from the literature (see Chapter 2), those that may confound the association between age of opportunity to use cannabis and later outcomes were identified through multivariate logistic regression. Covariates were included in the logistic models if significantly

Figure 3.4: Logistic regression analyses for age of cannabis opportunity and later cannabis use outcomes of daily use, abuse and/or dependence, and treatment seeking. Covariates identified for adjusted analyses listed in boxes above and below the independent variable



associated with both age of opportunity (see Table 3.4) and the dependent variable (see Appendix 3).

3.5.2 Results

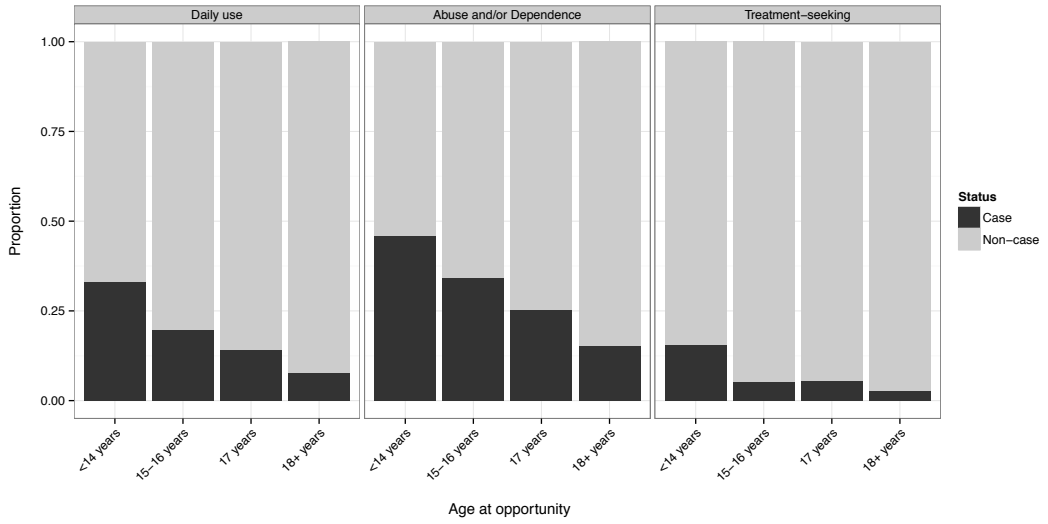
Analysis of the Relationship between Age of Opportunity to Use Cannabis and Cannabis Outcomes of Daily Use, Abuse and/or Dependence, and Treatment Seeking

Significant differences were observed between the age of opportunity groups for the prevalence of daily cannabis use, cannabis abuse and/or dependence and cannabis related treatment seeking (see Table 3.3). Amongst those who reported opportunity to use cannabis at age 14, daily cannabis use (28.1%), cannabis abuse and/or dependence (38.9%), and treatment seeking (13.1%). Figure 3.5 demonstrates that the proportion of those reporting these outcomes decreased as age of opportunity to use cannabis increased, and indicates a linear relationship between transition speed and all later outcomes.

Table 3.3: Association between age of first opportunity to use cannabis and daily cannabis use, cannabis abuse and/or dependence, and cannabis related treatment seeking in the ATR sample (N=3399)

Variable	Under 14 N = 388	15-16 N = 986	17 N = 558	18 and over N = 1467	Phi coefficient	χ^2 P value
	N (%)	N (%)	N (%)	N (%)		
Daily cannabis use N=372	109 (28.1)	149 (15.1)	54 (9.7)	60 (4.1)	0.25	$\leq$**0.001**
Cannabis abuse and/or dependence N=624	151 (38.9)	259 (26.3)	96 (17.2)	119 (8.11)	0.27	$\leq$**0.001**
Treatment seeking N=132	51 (13.1)	39 (4.0)	21 (3.8)	22 (1.5)	0.18	$\leq$**0.001**

Figure 3.5: Diagram visualising the prevalence of cannabis outcomes by age of opportunity to use cannabis



Association Between Age of Opportunity to Use Cannabis and Potential Model Covariates

After adjustment for other variables within the model, a number of factors were identified as significantly associated with age of opportunity to use cannabis analysis groups (see Table 3.4). Gender, education, CD, depressive episode, parent drug problems, strict parenting, high levels of school peer cannabis use, infrequent childhood religious attendance, lifetime monthly alcohol use, lifetime weekly tobacco use, lifetime other drug use, lifetime alcohol dependence and lifetime drug dependence were all associated with age of opportunity to use cannabis.

Table 3.4: Association between age of first opportunity to use cannabis and socio-demographic, childhood, mental health, peer and drug use factors in the ATR sample (N=3399)

Variable	Age at first cannabis use opportunity (years)			
	Under 14 N = 388	15-16 N = 986	17 N = 558	18+ N = 1467
	N (%) OR (95% CI)	N (%) OR (95% CI)	N (%) OR (95% CI)	N (%) OR (95% CI)
Male gender	166 (42.8) 1.24 (0.94-1.64)	428 (43.4) **1.26*** (1.04-1.53)	222 (39.8) 1.20 (0.97-1.49)	483 (32.9) 1.0
Lower level of completed education	123 (31.7) 1.26 (0.94-1.69)	280 (28.4) **1.30**** (1.06-1.61)	147 (26.3) **1.30*** (1.02-1.65)	290 (19.8) 1.0
Conduct disorder	114 (36.1) **6.22***** (4.08-9.47)	129 (40.8) **3.37***** (2.25-5.05)	35 (6.3) **1.74*** (1.05-2.87)	38 (2.6) 1.0
Non-clinical depressive episode	220 (57.1) 0.87 (0.66-1.14)	448 (45.7) **0.77**** (0.64-0.92)	259 (46.4) 0.88 (0.71-1.08)	710 (48.5) 1.0
Parental alcohol problems	134 (34.5) 1.14 (0.84-1.54)	271 (27.5) 1.06 (0.85-1.32)	140 (25.1) 0.96 (0.75-1.24)	351 (23.9) 1.0
Parental drug problems	36 (9.3) **2.09**** (1.18-3.69)	39 (4.0) 1.29 (0.79-2.10)	17 (3.1) 1.12 (0.63-2.01)	34 (2.3) 1.0
Single parent family	39 (10.1) 1.53 (0.87-2.69)	55 (5.6) 0.95 (0.62-1.45)	37 (6.6) 1.30 (0.84-2.02)	72 (4.9) 1.0
High parental conflict	170 (13.3) 1.18 (0.88-1.57)	368 (28.9) 1.02 (0.84-1.24)	209 (16.4) 1.12 (0.90-1.40)	526 (41.3) 1.0
Strict parenting	207 (53.4) 0.87 (0.67-1.13)	470 (47.8) **0.81*** (0.68-0.98)	254 (45.5) **0.78*** (0.63-0.96)	741 (50.6) 1.0
High levels of peer cannabis use	75 (19.3) **6.78***** (4.20-10.97)	99 (10.0) **3.77***** (2.45-5.82)	31 (5.6) **2.25***** (1.38-3.67)	36 (2.5) 1.0
Childhood sexual abuse	62 (16.3) 1.48 (0.97-2.26)	82 (8.4) 0.99 (0.70-1.40)	44 (7.9) 1.05 (0.73-1.52)	105 (7.2) 1.0
Infrequent childhood religious attendance	195 (50.3) **1.81***** (1.38-2.37)	465 (47.2) **1.66***** (1.38-1.99)	238 (42.7) **1.36***** (1.10-1.67)	520 (35.5) 1.0
Lifetime monthly alcohol use	366 (94.3) 0.66 (0.38-1.14)	925 (93.8) **0.70*** (0.49-0.99)	526 (94.3) 0.94 (0.61-1.46)	1366 (93.1) 1.0
Lifetime weekly tobacco use	257 (66.4) **2.21***** (1.56-3.13)	526 (53.4) **1.86***** (1.47-2.36)	250 (44.8) **1.52***** (1.16-2.00)	461 (31.4) 1.0
Lifetime other drug use	281 (72.4) **2.26***** (1.71-3.00)	551 (55.9) **1.53***** (1.27-1.85)	257 (46.1) 1.19 (0.95-1.49)	535 (36.5) 1.0
Lifetime alcohol dependence	165 (42.5) **1.78***** (1.34-2.38)	337 (34.2) **1.80***** (1.46-2.22)	161 (28.9) **1.63***** (1.26-2.10)	265 (18.1) 1.0
Lifetime tobacco dependence	184 (47.4) 1.21 (0.84-1.75)	342 (34.7) 1.12 (0.85-1.47)	154 (27.6) 1.06 (0.77-1.46)	267 (18.2) 1.0
Lifetime other drug dependence	60 (15.5) **1.95**** (1.19-3.20)	62 (6.3) 1.22 (0.78-1.90)	24 (4.3) 1.17 (0.67-2.04)	33 (2.3) 1.0

*P $\leq$0.05 **P$\leq$0.01 ***P $\leq$0.001.

Adjusted Analysis of the Relationship Between age of Opportunity and Later Cannabis Outcomes

The results of the logistic regression model (see Table 3.5) show that progression to daily cannabis use is more than twice as likely amongst those who have the

earliest opportunity to use cannabis (OR 2.06, 95% CI 1.36-3.11), and more likely amongst those who were age 15-16 (OR 1.67, 95% CI 1.18-2.37) when they had their first opportunity to use cannabis, compared to those who were 18 and over. Reporting cannabis abuse and/or dependence was more likely amongst who were under 14 (OR 1.91, 95% CI 1.33-2.74) or 15-16 (OR 1.86, 95% CI 1.40-2.48), and aged 17 (OR 1.51, 95% CI 1.08-2.11). Significant associations with treatment seeking were only observed for those who had their first opportunity to use cannabis before the age of 14. This group were more than twice as likely to progress to seeking treatment for cannabis use (OR 2.65, 95% CI 1.51-4.64). Post hoc tests revealed differences between levels were not significant.

Table 3.5: Odds ratios (95% Confidence intervals) for association between age of opportunity to use cannabis, covariates and later cannabis outcomes from logistic regression analysis of the ATR sample (N=3399)

Age at Opportunity to Use Cannabis	Daily Use N = 372 Odds Ratio (95% Confidence) (Interval)		Abuse and/or Dependence N = 624 Odds Ratio (95% Confidence) (Interval)		Treatment Seeking N=132 Odds Ratio (95% Confidence) (Interval)	
	Univariate model	Adjusted model	Univariate model	Adjusted model	Univariate model	Adjusted model
Under 14 N=388	**9.16*** ** (6.46 - 12.99)	**2.06*** ** (1.36-3.11)	**7.22*** ** (5.40 - 9.65)	**1.91*** ** (1.33-2.74)	**9.94*** ** (5.96 - 16.59)	**2.65*** ** (1.51-4.64)
15–16 N=986	**4.17*** ** (3.05 - 5.71)	**1.67*** ** (1.18-2.37)	**4.04*** ** (3.16 - 5.15)	**1.86*** ** (1.40-2.48)	**2.70*** ** (1.59 - 4.61)	1.14 (0.65-2.00)
17 N=558	**2.51*** ** (1.70 - 3.71)	1.51 (0.97-2.34)	**2.35*** ** (1.75 - 3.16)	**1.51*** (1.08-2.11)	**2.57** ** (1.35 - 4.89)	1.57 (0.81-3.07)
18 and over N=1467	1	1	1	1	1	1
Covariate						
Male gender	**1.54*** ** (1.18-2.02)		**1.89*** ** (1.51-2.36)		1.38 (0.93-2.04)	
Lower level of completed education	**1.65*** ** (1.25-2.18)		**1.38** ** (1.09-1.75)		1.31 (0.90-1.90)	
Conduct disorder	**2.19*** ** (1.58-3.02)		**2.23*** ** (1.64-3.03)		-	
Lifetime weekly tobacco use	**4.62*** ** (3.27-6.51)		**2.99*** ** (2.35-3.81)		**3.36*** ** (1.86-6.07)	
Lifetime other drug use	**6.32*** ** (4.12 - 9.70)		**4.32*** ** (3.28-5.69)		**3.93*** ** (2.03-7.60)	
Lifetime other drug dependence	**2.72*** ** (1.87-3.95)		3.18 (2.18-4.64)		**3.91*** ** (2.48-6.16)	

*P ≤0.05 **P≤0.01 ***P ≤0.001.
All models have applied the Huber-White adjustment for family clustering

Results Summary 2

Age of opportunity to use cannabis under age 18 was associated with lifetime daily cannabis use and cannabis abuse/or dependence, with effect sizes indicating a linear relationship between younger age of opportunity and increased likelihood of these outcomes.

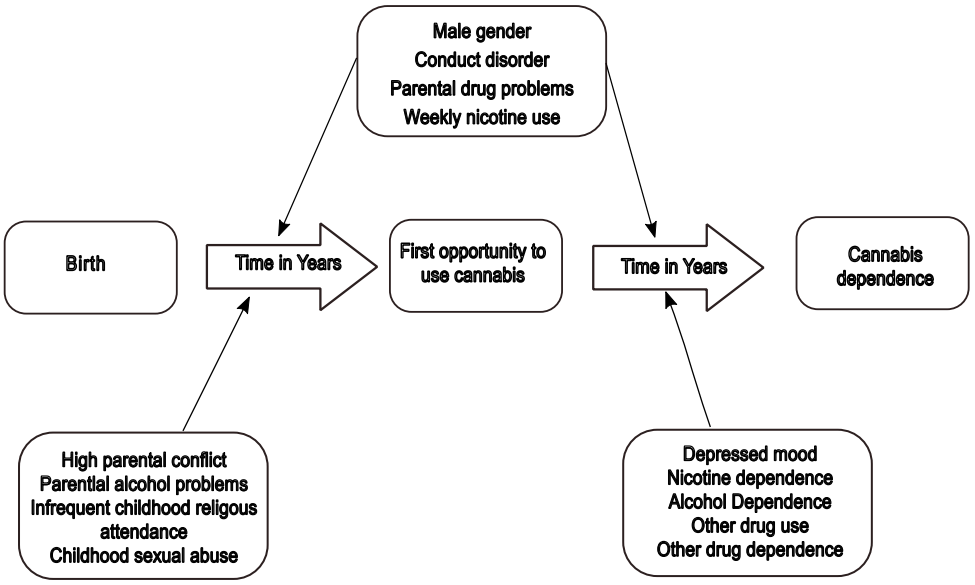
3.6 Discussion

3.6.1 Summary

A number of factors were uniquely associated with the transition to cannabis use opportunity, and with the transition from opportunity to cannabis dependence, but several factors were found that increase hazards of both these transitions. Parental conflict, parental alcohol problems, infrequent childhood religious attendance and CSA were uniquely associated with faster transition to opportunity. Depressive episode, tobacco dependence, alcohol dependence, other drug use and other drug dependence were uniquely associated with faster progression from opportunity to dependence. CD, parental drug problems, male gender and weekly tobacco use were associated with faster progression to both opportunity and from opportunity and dependence. See Figure 3.6 for an overview of these findings.

Given the overlapping influences, it is perhaps unsurprising that the age of opportunity to use cannabis was found to be significantly associated with later cannabis use outcomes. Those whose first opportunity to use cannabis occurred before the age of 14 were more than twice as likely to experience daily cannabis use and cannabis related treatment seeking, and almost twice as likely to experience cannabis abuse and/or dependence, compared to those reporting cannabis opportunity after age 18. Those who reported cannabis use opportunity age 15-16 were almost twice as likely to report daily cannabis use and cannabis abuse and/or dependence, and those who were age 17 at opportunity to use cannabis age 17 were almost twice as likely to report cannabis abuse and/or dependence, compared to those reporting cannabis use opportunity after age 18.

Figure 3.6: Unique and consistent associations across the transitions 1) birth to opportunity to se cannabis 2) opportunity to use cannabis to the development of dependence



3.6.2 Factors Associated With Multi-Stage Transitions

Survival analysis identified a number of factors associated with both transitions. Male gender was associated with faster progression to both opportunity and dependence. In contrast to previous research, gender differences held across both transitions. Previous research has found males more likely to have *lifetime* opportunity to use cannabis, but that these gender differences do not extend to the transition into drug use once opportunity has occurred (van Etten et al., 1999). Similarly, weekly tobacco use was associated with increased hazard of both cannabis use opportunity and progression to cannabis dependence, consistent with existing findings relating to use (Wagner and Anthony, 2002). CD was associated with faster progression to both opportunity and dependence, echoing previous research showing disruptive or aggressive behaviour in both males and females is associated with earlier opportunity to use cannabis (Storr et al., 2011), and that CD increases risks of the development of cannabis dependence (Agosti et al., 2002; Elkins et al., 2007).

Parental drug problems were significantly associated with a more rapid transition to both opportunity and dependence, in line with existing research demonstrating increased cannabis use opportunity amongst those with parental

drug problems (Benjet et al., 2013). Given the especially strong association with opportunity to use cannabis it is plausible that parental drug problems facilitate an environment in which drug access is increased, whether this is indirectly or directly through parents.

The identification of tobacco, alcohol and other drug involvement as factors associated with progression to dependence suggests that a pattern of poly-use emerges. Although alcohol use has previously been found to be associated with early onset of cannabis use (Coffey et al., 2000) it was not associated with opportunity to use cannabis in the present analyses. The comparatively rarer outcomes of tobacco dependence, other drug use and other drug dependence were found to be associated with increased speed of progression to cannabis dependence. The co-occurrence of tobacco and cannabis use has been frequently observed (Agrawal et al., 2010; Hindocha et al., 2015, 2016), and regular cigarette smokers are more likely to report earlier cannabis use opportunity (Agrawal et al., 2013). Present results strongly supported this finding, and extend it to show weekly tobacco use and dependence were significantly associated with speed of progression to cannabis dependence. The observed association between cannabis dependence and tobacco may be due to a number of factors including shared genetic and environmental influences, the co-administration of tobacco and cannabis, and smoking habituation (Agrawal et al., 2012).

3.6.3 Age Of Opportunity To Use Cannabis And Later Outcomes

The results demonstrate a linear relationship between speed of transition to opportunity to use cannabis (represented through earlier age at first opportunity) and the outcomes of daily use and abuse and/or dependence. This is a novel finding as the relationship between age of opportunity and later outcomes has not been previously explored in the literature (see Chapter 1). The findings are interesting given that opportunity to use cannabis does not incorporate psychopharmacological elements of cannabis use on the developing brain, which could be theorised to explain the association between early initiation and later dependence. Although the mechanisms of this have not been explored in relation to later cannabis use outcomes, those who initiate cannabis use earlier have been found to experience attentional and working memory dysfunctions, demonstrated

through cortical activation compared to alter onset users and non-users (Becker et al., 2010; Kempel et al., 2003; Skosnik et al., 2006). The present results suggest that the association between early initiation and later dependence may be separate from psychopharmacology.

Given the associations between early initiation of cannabis and later problematic cannabis use (Swift et al., 2008; Grant and Dawson, 1998; Fergusson and Horwood, 1997), this may suggest that age of opportunity to use cannabis is a proxy for age of cannabis initiation, and an earlier point on the same liability axis. Within the sample the majority of participants within each age group progressed to initiation of cannabis use in less than a year after having had the opportunity to use cannabis (analyses not included in thesis due to homogenous speed of transition; mean = 0.74 years, s.d. = 1.91, range = 0 - 19). This is slightly faster than published estimates of the median delay between opportunity and use of a drug, which was estimated at 1 year (Swendsen et al., 2008). The effect sizes obtained for the relationship between age of opportunity and cannabis abuse and/or dependence in the present analysis broadly map onto the effect sizes obtained in a study of the relationship between age of cannabis initiation and DSM-III marijuana disorder (DeWit et al., 2000), which found that in comparison to those who initiated age 20 and over likelihood of dependence symptoms was increased in those who initiated use age 14 (OR 2.28, CI 1.34 – 3.97, compared to present finding of OR 1.91 95% CI 1.33-2.74 for opportunity age 14 and under), 16 (OR 1.93, CI 1.19 – 3.20, compared to present finding of OR 1.86 95% CI 1.40 – 2.48 for opportunity age 15-16), and 17 (OR 1.78, CI 1.06 – 3.04, compared to present finding of OR 1.51, 95% CI 1.08 – 2.11 for opportunity age 17).

The associations between earlier age of opportunity to use cannabis and later cannabis-related outcomes remained (albeit with a large decrease in effect size) after controlling for the identified potentially confounding variables. This suggests that other factors may be underlying this association. It is plausible that some of the same genetic factors are underlying both opportunity to use cannabis and cannabis dependence. This will be studied in the next chapter.

3.6.4 Limitations

There are certain considerations required in interpretation of this work. Firstly, analyses were conducted on retrospective self-report data, introducing the possibility of recall bias. This is a viable method of data collection (Darke, 1998), and indeed recall of early experience with cannabis has been found to be especially reliable (Johnson and Mott, 2001), but as the analyses rely on accurate recall of age of onset of a number of behaviours the work would benefit from replication in longitudinal cohorts. Secondly, analyses of the progression from opportunity to cannabis use initiation were not possible, as timing of transitions was only available as time in years, and there was not enough variation in the speed of this transition to allow for analysis (the majority of participants progressed to use within 1 year after having the opportunity to use, data available on request). Thirdly, selected covariates measured occurrence within an age range (6-13), and consequently may have occurred prior to the age of opportunity to use cannabis for a small number of individuals. Fourthly, while the prevalence of lifetime cannabis use in this sample was relatively high at 68.2% it is important to note this estimate is consistent with previous estimates from the Australian young adult population (of Health and Welfare, 2014). Finally, interpretation of these analyses should be in light of the twin and sibling sample used, as there is some residual uncertainty about whether inferences from twin data have external validity with respect to what might be found in general population samples (Vitaro et al., 2009). Analyses were adjusted for clustering effects using the Huber-White estimator, which was selected over other potential analyses that can be conducted to explore within twin/sibling frailties as the most parsimonious method.

3.6.5 Implcaitions

Consideration of multiple stages of drug use from non-use to dependence allows identification of factors uniquely associated with specific transitions. The current results demonstrate that different factors are influential at different stages of the development of cannabis dependence. Additionally, the differences and consistencies in factors across the stages of drug use provide an insight into which similarities and differences we may expect to see occurring through the transitions towards dependence. The findings have implications for substance use

prevention efforts, as both the targeting of interventions as well as the interventions themselves may benefit from being tailored to stages of drug use. Speculatively, adolescents may feel more comfortable reporting having had the opportunity to use cannabis then initaition of cannabis, as opportunit does not involve reporting illegal activity. It may therefore prove a useful tool for targeting interventions.

CHAPTER 4

Go on Until you Reach the End: Genetic Correlation Between Cannabis Opportunity and Abuse and/or Dependence

4.1 Introduction

In the previous chapter an association was observed between age of opportunity to use cannabis and the development of abuse and/or dependence. This association remained after adjusting for the effect of factors that were associated with progression to both stages. It is possible that shared genetic factors may underlie part of the association between these two variables. Cannabis use disorders have a strong genetic component, with a review of 6 studies in the area concluding heritability estimates range from 45% – 78% (Agrawal and Lynskey, 2006). Opportunity to use a drug may be a putatively environmental factor, but such factors have been shown to have heritable influences (Kendler and Baker, 2007). Heritable influences on early stages of cannabis use, such as the opportunity to use cannabis, may include personality factors and behaviours which have been shown to be associated with earlier exposure (such as disruptive childhood behaviour

(Storr et al., 2011)) and are influenced by additive genetic influences (Hudziak et al., 2003). In contrast, genetic influences on abuse or dependence can be hypothesised to include factors associated with drug metabolism.

Heritable and environmental influences on progression through stages of drug use may vary (see Chapter 1), and some risk factors may be unique to specific stages of substance involvement, while others may act, or be correlated, across multiple stages (Kendler et al., 1999a). No research to date has explored whether there is overlap between factors associated with earlier opportunity and those associated with lifetime cannabis abuse and/or dependence.

Twin studies estimate additive genetic effects, common environment effects and unique environment effects on a phenotype (Neale and Cardon, 1992). Two-stage bivariate twin modelling (Heath et al., 2002) is an extension of bivariate twin modelling. This model is used to estimate the correlation between additive genetic (A), shared environment (C) and unique environment (E) in situations where early-stage phenotypes, such as opportunity to use a drug, are *necessary* for the expression of later behaviours, such as abuse and/or dependence. In order to overcome the issue that those who have not had the opportunity to use cannabis will have structural missing data for the outcome of dependence, the initial variable of opportunity is coded to have at least three categories, two of which include individuals who can be assessed on the second outcome.

The present study aims to apply bivariate models to twin data on cannabis opportunity and abuse and/or dependence to determine the extent to which genetic influences on the development of cannabis abuse/dependence are unique to the phenotype, and the extent to which they are correlated with genetic influences on opportunity to use cannabis.

Aim

Establish the extent to which age of opportunity to use cannabis is influenced by additive genetic, shared environmental and unique environmental factors, and the extent to which these influences correlate with those influencing the development of abuse and/or dependence.

Hypothesis 1

There will be genetic influences on age of opportunity to use cannabis, and on lifetime cannabis abuse and/or dependence.

Hypothesis 2

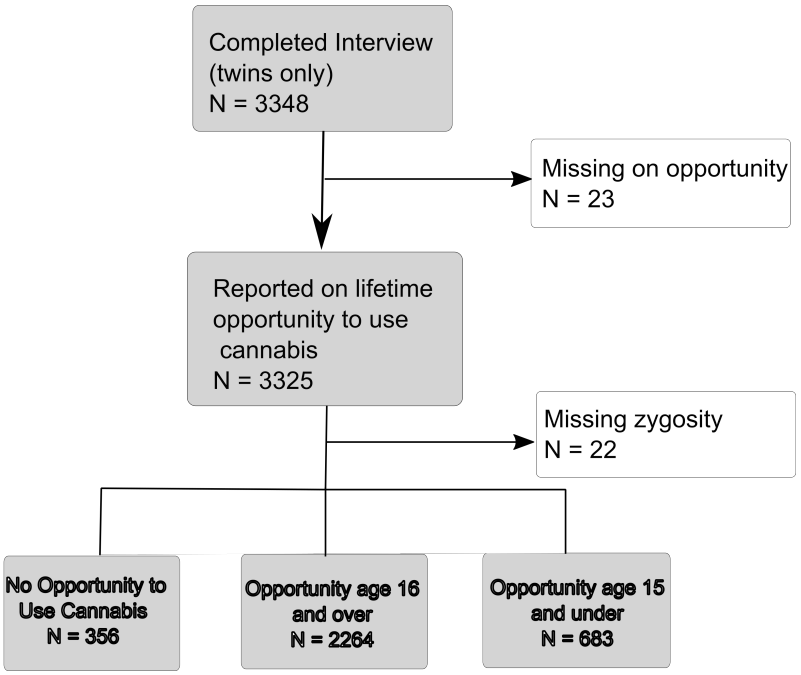
Genetic influences on age of opportunity to use cannabis will be moderately correlated with those influencing the development of cannabis abuse and/or dependence.

4.2 Methods

4.2.1 Sample

The sample was drawn from the Australian Twin Registry (ATR). The 3303 twins who provided information on whether or not they had ever had the opportunity to use cannabis, and who had complete zygosity information, form the analysis sample in this chapter (see Figure 4.1).

Figure 4.1: ATR sample for analyses in this chapter



4.2.2 Measures

For these analyses age of opportunity was coded into three levels. These were having never had the opportunity to use cannabis (N = 356), having had later

opportunity to use cannabis (first opportunity reported as happening at age 16 and over, N = 2264), or having had early opportunity to use cannabis (first opportunity reported as happening at age 15 or earlier, N = 683). The variable levels were coded in this order to represent the increasing liability to the outcome associated with each level. Sensitivity testing of the cut-off of 15 and under for early cannabis use opportunity is available in Appendix 5. The correlations obtained by different cut-off points indicated that results were not affected by the choice of 15 as age cut-off for early opportunity (see Appendix 5).

Cannabis abuse and/or dependence was derived from participants meeting the DSM-IV criteria for either of these disorders (American Psychiatric Association, 2000) (derivation outlined in Chapter 2 Section2.2.2). Within the twin only subsample that had provided data on opportunity to use cannabis, 16.5% (N = 547) participants reported meeting criteria for cannabis abuse and/or dependence.

4.3 Analytic Strategy

4.3.1 Testing the Assumptions of Common Liabilities

In order to establish if there were shared liabilities between lifetime opportunity to use cannabis (coded yes/no for this part of the analysis) and abuse/dependence a Causal Common Contingent model was used. This provides a beta pathway between the two phenotypes, which suggests correlated liabilities if significant (Do et al., 2015), and if the beta pathway is greater than 0 (independent liability distribution) but less than 1 (single liability distribution) (Maes et al., 2004b). A significant beta test indicating moderate to high multiple liabilities (common causes underlying both phenotypes) demonstrates that there are shared influences acting on both phenotypes, and further bivariate twin modelling is necessary to determine the sources of these common causes.

4.3.2 Equating Thresholds and Correlations

In order to test whether thresholds and correlations could be equated between MZ and DZ males and females, a series of nested models were compared against a complete saturated model (see Chapter 2, Section 2.5.5 for full explanation of

nested modelling). If the nested model was not significantly different ($P \leq 0.05$) to the saturated model then the parameters tested were equated, in order to develop the most parsimonious model.

4.3.3 Two-Stage Bivariate Twin Model

A two-stage twin model was fitted to assess contributions of additive genetic (A), shared environmental (C), and unique environmental (E) factors to the variance in age of opportunity to use cannabis and on lifetime cannabis abuse and/or dependence, and to estimate the extent to which influence of A, C and E on the two phenotypes were correlated (Heath et al., 2002). This model is a variation of the classic bivariate models that is appropriate for analysis of variables with data missing at random (data that are missing as a result of observations on a previous variable, as opposed to data that are missing completely at random (Little and Rubin, 2014)). In the present analysis twins who had no opportunity to use cannabis have a 100% a priori probability of having a value of zero for the outcome cannabis abuse/dependence, resulting in structural missing data. The model coded opportunity as a three level variable (never opportunity/later opportunity/early opportunity, as described in the measures section above), two levels of which have non-missing data on abuse/dependence. A constrained correlations model is applied, which utilises the cross-covariance within twin pairs, which refers to the correlation between trait X in twin 1 and trait Y in twin 2 (Plomin et al., 2013).

4.3.4 Sex-Limitation Bivariate Modelling

Sex limitation techniques can be applied to multivariate modelling to determine whether the effects of genetic (or environmental) factors differ between males and females. Sex differences can be scalar, in which the same factors affect the phenotype in both sexes but the magnitude of the effect differs, or non-scalar, in which one or more factors affect a phenotype in one sex but not the other (Neale et al., 2006c). Scalar sex differences can be assessed through the covariance between same-sex MZ and DZ twins, whereas assessment of non-scalar sex differences require the inclusion of opposite-sex DZ twins to assess whether correlation within these pairs is reduced compared to same-sex DZ twin pairs.

Caution needs to be taken when applying sex limitation modelling in a multivariate twin analysis context. In an in-depth exploration of the issue, Neale et al. (2006c) tested the optimum way to approach the analysis, outlining a number of requirements for the generation of covariance matrices within this analysis. These include:

1. The processes causing individual differences must cause positive variance in the phenotypes;
2. Correlation matrices must remain consistent with each other (e.g. if the correlation between X and Y is 1.0, the correlation between X and Z must equal the correlation between Y and Z);
3. The predicted phenotypic covariance must be positive definite (each of the values contributing to the equation must be positive).

4.3.5 Selection of Correlation Model over Cholesky Decomposition

Bivariate twin modelling commonly utilises a Cholesky Decomposition analysis, whereby variance in the first variable in the model is assumed to be caused be a latent factor that can explain the variance in the second variable. The variance in the second variable in the analysis is assumed to be caused by a second latent variable, which does not explain the variance in the first variable. Consequently all variables load on to the first latent factor, but only the second variable in the analysis will load on to the second latent factor (Gillespie and Martin, 2005). This allows estimation of the extent to which two variables share latent factors.

For scalar sex limitation, an additional requirement is that the factors must covary to the same extent in males and females, resulting in a common factor correlation structure with different factor loadings for males and females. As a result, the use of Cholesky Decomposition analysis has been found to be unsuitable for sex limitation models, as it does not retain the required constraint for scalar sex limitation that the factors correlate equally in males and females(Neale et al., 2006c).

A correlation approach to the multivariate sex-limitation model has been found to be more suitable. The correlation model can be specified in terms of factors for males and females, and can estimate covariance within male and female factors separately. These factors are constrained to have the same covariance matrix in

males and females. The factors then influence the phenotype via path coefficients that are not constrained to be equal across sexes. Thus the model specifies that the same factors influence males and females but that they may do so to different degrees. Path analysis can then be conducted on the correlated model to determine the variance unique to each factor, and the variance shared between factors, providing the same estimates that are achieved with a Cholesky model.

4.3.6 Analysis Summary

First, the assumption of correlated liabilities underlying both phenotypes was tested using a CCC model to estimate the beta pathway. A liability threshold model estimating co-twin correlations was fitted to the data set and used to test assumptions regarding the equality of thresholds between sexes, and MZ and DZ correlations. Next, a constrained correlation bivariate model was fitted, partitioning the variance attributable to A C and E for both phenotypes, and the A C and E correlations between these variance components. A freely estimated model was fitted for both sexes, and then separate nested models fitted to test whether parameters and correlations could be equated across sexes. Difference in model fit was assessed via the likelihood-ratio χ^2 test and examination of the Akaike Information Criterion (AIC) (a description of these tests of model fit is provided in Chapter 2, Section 2.5.5).

4.4 Results

4.4.1 Prevalence of, and Correlations between, Opportunity to use Cannabis and Abuse and/or Dependence

Of those who reported opportunity to use cannabis by age 15, 32.9% (N = 191) reported cannabis abuse and/or dependence. Of those who reported cannabis use opportunity at age 16 or older, 13.7% (N = 269) reported cannabis abuse and/or dependence. A saturated model was used to estimate tetrachoric correlations for the traits of age of opportunity and lifetime cannabis abuse and/or dependence (see table 1). The phenotypic correlation was 0.44 in males and 0.44 in females. The across twin/across trait correlation between age of opportunity to use cannabis

and cannabis abuse and/or dependence (see Table 4.1) was similar in MZ and DZ males but almost twice as high in MZ female pairs compared to DZ female pairs, indicating there may be stronger influence of additive genetic effects on the correlation in females.

Table 4.1: Phenotype correlations for ATR MZ and DZ male, female and opposite sex twin pairs (N=3303)

	Opportunity/Abuse and/or dependence twin 1/twin 2		Age of opportunity twin 1/twin 2		Abuse and/or dependence twin 1/twin 2	
	Correlation	95% CI	Correlation	95% CI	Correlation	95% CI
MZ males N=481	0.41	(0.26 – 0.54)	0.72	(0.57 – 0.83)	0.72	(0.52 – 0.85)
DZ males N=371	0.47	(0.46 – 0.63)	0.48	(0.22 – 0.68)	0.46	(0.16 – 0.69)
MZ females N=975	0.42	(0.30 – 0.53)	0.63	(0.54 – 0.71)	0.83	(0.71 – 0.85)
DZ females N=734	0.17	(0.02 – 0.31)	0.42	(0.28 – 0.54)	0.48	(0.39 – 0.63)
DZ opposite sex N=742	0.23	(0.06 – 0.37)	0.26	(0.09 – 0.42)	0.33	(0.07 – 0.37)

4.4.2 Assumption Testing

Results of CCC modelling indicated significant correlated liabilities between cannabis opportunity and the development of cannabis dependence, with the beta estimated at 0.72 (95% CI 0.71 – 0.72). This indicates that there is correlation in the factors underlying expression of the two traits.

Thresholds for age of opportunity to use cannabis and for cannabis abuse and/or dependence could not be equated (for both models tested against the saturated model, P=$\leq$0.001). Within trait and within twin correlations could be equated for MZ male and female twins, DZ male and female twins, and DZ M/F twins and OS twin pairs with no significant loss of model fit (compared to saturated model Δ-2LL=12.7, ΔDF= 9, P value = 0.18). This precluded the necessity to test for non-scalar sex differences (Neale et al., 2006c). See Table 4.2 for full model comparison statistics.

Table 4.2: Testing whether thresholds and correlations can be equated between male and female twins (N=3303)

Model number	Model name	Comparison model	-2LL	DF	AIC	Δ-2LL	ΔDF	P value
1	Saturated model	-	7685.9	6582	-5478	-	-	-
2	Equating m/f opportunity thresholds	1	7760.2	6584	-5407	74.3	2	≤**0.001**
3	Equating m/f abuse and/or dependence thresholds	1	7779.5	6583	-5386.6	93.6	1	≤**0.001**
4	Equating MZ m/f within trait and and within twin correlations	1	7688.4	6585	-5481.6	2.5	3	0.47
5	Equating DZ male/female within trait and within twin correlations (MZ MF correlations equated)	4	7695.4	6588	-5480.7	6.9	3	0.07
6	Equating DZ male/female and OS twin within trait and within twin correlations (DZ MF and MZ MF correlations equated)	5	7698.6	6591	-5483.4	3.2	3	0.36
7	Correlations equated for all MZ and DZ twin pairs; m/f thresholds freely estimated	1	7698.6	6591	-5483.4	12.7	9	0.18

The correlations for a model where male and female correlations were equated for MZ pairs were:

- Opportunity/Abuse and/or dependence twin 1/twin 2: 0.45 (95% CI 0.38 - 0.52)
- Opportunity twin 1/twin 2: 0.65 (95% CI 0.58 - 0.72)
- Abuse and/or dependence twin 1/twin 2: 0.41 (95% CI 0.32 - 0.48)

The correlations for a model where male, female and opposite sex correlations were equated for DZ pairs were:

- Opportunity/Abuse and/or dependence twin 1/twin 2: 0.26 (95% CI 0.16 - 0.35)
- Opportunity twin 1/twin 2: 0.37 (95% CI 0.27 - 0.46)
- Abuse and/or dependence twin 1/twin 2: 0.41 (95% CI 0.26 - 0.44)

As converting continuous variables to categorical can be a source of bias in analyses (Altman, 1994), sensitivity analyses were conducted for the early opportunity to use cannabis cut-off age. The analysis indicated that correlations obtained did not differ by choice of the age cut-off (see Appendix 5).

Figure 4.2: ACE parameter and correlation estimates for females (freely-estimated correlation model) (N=1709)

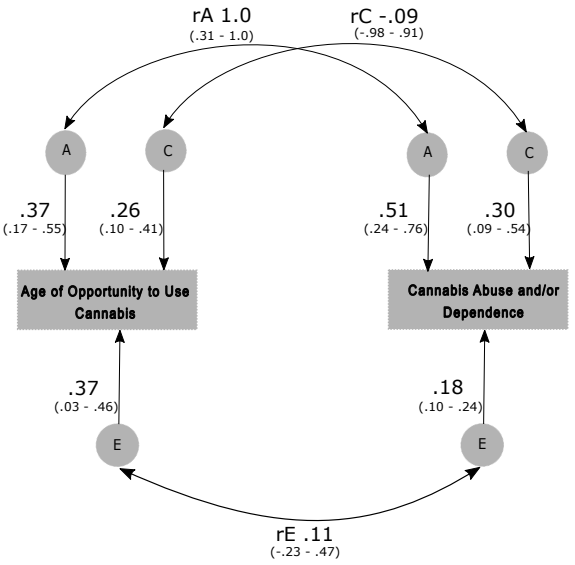
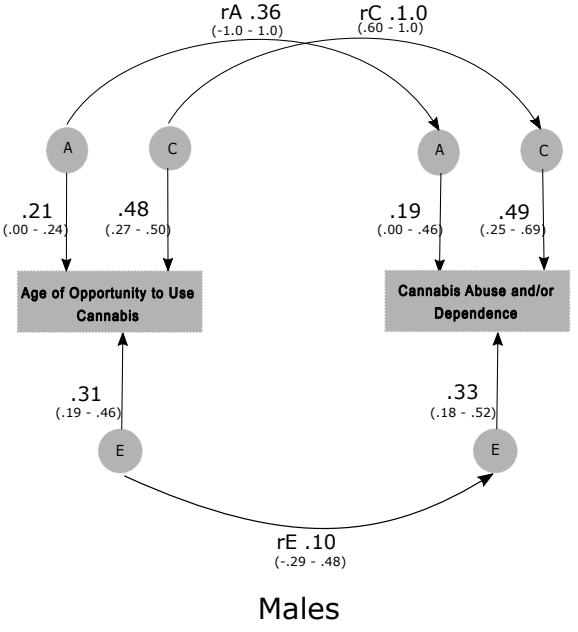


Figure 4.3: ACE parameter and correlation estimates for males (freely-estimated correlation model) (N=852)



4.4.3 Constrained Correlations Bivariate Sex Limitation Model Fitting

The results of the full freely-estimated correlation model, with separate parameter and correlation estimates for males and females, are provided in Figure 4.2 and

Figure 4.3. The fit statistics for this model were -2LL=7690.6, DF=6585, AIC=-5479.4.

4.4.4 Nesting Models to Develop Parsimonious Model Fit

In order to achieve the most parsimonious model fit, nested models were fitted equating the A, C and E variance components and correlations for males and females. It was possible to equate the estimates for A (-2LL=7694.0, DF=6588, AIC=-5482.0, compared to freely estimated model Δ-2LL= 3.5, ΔDF= 3, P value = 0.33), C (-2LL=7696.0, DF=6588, AIC=-5480.0, compared to freely estimated model Δ-2LL=5.4, ΔDF= 3, P value = 0.15), and E (-2LL=7695.1, DF=6588, AIC=-5480.9, compared to freely estimated model Δ-2LL= 4.5, ΔDF= 3, P value = 0.21). A model equating ACE simultaneously across males and females was not a significantly worse model fit compared to the freely estimated model (-2LL=7698.9, DF=6594, AIC=-5489.2, compared to freely estimated model Δ-2LL=8.3, ΔDF= 9, P value = 0.51). The contribution of C to this fully equated model was found to be small (0.08 for age of opportunity, 0.04 for abuse and/or dependence). Consequently, the fit of a model where C was dropped was tested against the fit of the freely estimated model. The AE model did not have significantly worse fit than the freely estimated model (-2LL=7702.2, DF=6597, AIC=-5491.8, compared to freely estimated model Δ-2LL=11.6, ΔDF= 12, P value = 0.48).

4.4.5 Final Model: An AE Sex-Equated Model

As the AE model equated across males and females provided the most parsimonious model without significant deterioration of fit to the data, this was selected as the final model. Parameter estimates for age of opportunity to use cannabis were:

- A=0.66 (95% CI 0.59 – 0.70)
- E=0.34 (95% CI 0.28-0.41)

Parameter estimates for cannabis abuse and/or dependence were:

- A=0.78 (95% CI 0.68-0.86)

- E=0.22 (95% CI 0.14-0.32)

Correlations between the two phenotypes were estimated at:

- A = 0.59 (95% CI 0.48 – 0.70)
- E = -0.09 (95% CI -0.35 – 0.19)

Using path analysis, from these results it can be calculated that, of the variance in age of opportunity to use cannabis and cannabis abuse and/or dependence attributable to genetics, 42% is shared between these phenotypes ($\sqrt{0.66} \times \sqrt{0.78} \times 0.59$). From this, it can be estimated that 0.36 (0.78 - 0.42) (46%; 0.36÷0.78) of the additive genetic contribution to variance in cannabis abuse and/or dependence is unique to that phenotype.

4.5 Discussion

Additive genetic influences determine the majority of variance in both age of opportunity to use cannabis (0.66, 95% CI 0.59 – 0.70) and cannabis abuse and/or dependence (0.78, 95% CI 0.68-0.86). Of the additive genetic influences on cannabis abuse and/or dependence, 54% were common with genetic influences on opportunity, whilst 46% was specific to abuse and/or dependence. No effect of the shared environment on either age of opportunity to use cannabis or cannabis abuse and/or dependence was observed, but there were unique environmental influences on both phenotypes. Given that parameter estimates and correlations could be equated for males and females, there was no indication of scalar sex differences acting on these phenotypes.

Previous research has not explored the correlation between influences on cannabis use opportunity and cannabis abuse or dependence, but existing studies focussing on cannabis initiation have observed overlapping liabilities (0.88) to cannabis initiation and progression to heavy use (Fowler et al., 2007), and extremely high genetic correlation (r=.98) between frequency of cannabis use and cannabis dependence (Sartor et al., 2010). As such the present findings are in line with existing research showing genetic correlation between the early stages of cannabis use and later SUDs.

Opportunity to use cannabis is the necessary first step in progression towards problematic use, and is a phenotype that could be thought to be subject to external influence only,. However, there were moderate genetic influences on age of opportunity of cannabis use (0.66). Environmental measures can be heritable if an individual's behaviour impacts on the environmental exposures, and if aspects of that behaviour are subject to genetic influences (Kendler and Baker, 2007). A review of this area identified that positive and negative life events, divorce and social support all have heritable influences (Kendler and Baker, 2007). The previous chapter (Chapter 3) identified that CD influences transitions to cannabis use opportunity, and from opportunity to dependence. This is in line with existing research demonstrating the consistent influence of CD on drug use (Lynskey et al., 2002; Reboussin et al., 2015; Storr et al., 2011), and genes relating to CD and antisocial behaviour are plausible candidates that may contribute to the shared genetic liability between age of opportunity and the development of cannabis abuse and/or dependence.

Both cannabis opportunity and abuse and/or dependence show a moderate effect of the unique environment (0.34 and 0.22, respectively), but there is low negative correlation between the unique environmental influences on these phenotypes (r-0.09). This supports existing research demonstrating that the pattern of environmental factors associated with progression between specific stages of drug use differs between transitions (Belsky et al., 2013; Sartor et al., 2007); indeed, this was observed in the previous chapter (Chapter 5).

In the present analysis it was possible to drop C from the model without significant deterioration in model fit, indicating that none of the observed variance in opportunity to use cannabis or abuse and/or dependence in males was attributable to the shared environment in this sample. The shared environment is usually found to be more implicated at earlier stages than later (Fowler et al., 2007), These findings are contradictory to findings of a high C correlation between cannabis availability and cannabis abuse (Gillespie et al., 2007). The samples differ, with the Gillespie et al. (2007) findings based on an all-male population, but the findings may indicate that cannabis availability and opportunity are subtly different phenotypes.

4.5.1 Limitations

Certain limitations must be taken into account when interpreting these results. The data are based on retrospective self-report. Retrospective recall of age onset of drug use behaviours has been shown to be reliable (Ensminger et al., 2007)), but the analyses would benefit from replication in longitudinal cohorts. Self-report has been shown to be a valid measure of data collection relating to drug use (Darke, 1998), and has been described as the gold standard for collecting data on phenotypes such as initiation and opportunity (Wagner and Anthony, 2002). Given that use of cannabis is illegal (at time of data collection), some participants in the ATR study may have incorrectly reported on their drug use. However, given the high prevalence of self-reported lifetime cannabis use, it seems unlikely that this was a problem.

The results are based on a twin population. Research has demonstrated twin and non-twin populations do not differ in incidence of psychiatric illness (Kendler et al., 1996) and that no association has been found between twin environmental similarity and mental health outcomes (Kendler et al., 1993).

4.5.2 Implications

The findings of this paper have important implications for future studies of gene variants and heritability of problematic cannabis use, and in the choice of controls in case-control studies. These results indicate that only a moderate proportion of genetic influences on cannabis abuse and/or dependence are unique from those acting on age of opportunity to use cannabis. Such arguments have previously been made regarding the importance of considering drug use opportunity when looking at the genetics of opiate use. By comparing participants in treatment for opiate dependence with controls sourced from the ATR (individuals not dependent on alcohol or illicit drugs, with significantly lower illicit drug exposure), and separately with nondependent neighbourhood controls (high exposure to illicit drugs, either via use or from residing in environments with widespread drug availability), differences were found in the SNPs associated with heroin dependence (Nelson et al., 2013). Until now the importance for genetic studies of considering cannabis use opportunity has not been explored and no genetic studies of problematic cannabis use have considered cannabis use opportunity amongst the control

group. The present findings indicate that by not taking this into consideration, and by ignoring the stage-specific nature of drug use, there is a risk of conflation of genetic influences.

Consequently, a key implication of the current findings is the necessity of taking into consideration the stage of drug use reached amongst the controls. Existing research has utilised information on the extent of cannabis use in controls (e.g. excluding those who had used cannabis fewer than 6 times) (Hartman et al., 2009), but such issues are not always being taken into consideration (Benyamina et al., 2009). Depending on the research question, and on the development of research identifying genetic overlap between progression to other stages of cannabis use and problematic cannabis use, screening controls not only for opportunity or initiation of cannabis use, but also for frequency of use may have utility in improving cannabis dependence SNP identification in the future.

These findings may have further implications for of the overlap of genetic influences across drug classes. Existing research has suggested that a proportion of the genetic factors underlying SUDs are not specific to individual drugs, and that it is environmental influences that determine the drug of dependence (Kendler et al., 2003) However, previous research in this area has not incorporated consideration of the stage sequential nature of drug dependence into their analyses. Much of the non-specificity of genetic influences on SUDs may result from shared influences on the earlier stages of drug use, with more specific influences (such as those related to drug metabolism, for example) associated with later stages of use.

CHAPTER 5

A Curious Transition: Examination of Time from Initiation to Subsequent Use of Cannabis

5.1 Introduction

So far, the research in this thesis has focussed on the transition to opportunity to use cannabis. In this chapter I consider the transition from initiation to subsequent use of cannabis. This is a novel transition, previously unexplored in research (see Chapter 1), the speed of which is defined by the time between two stages. Studying transition speed that is defined by time between stages, rather than age of onset, removes the potential for the correlates of early initiation to mask associations that are specific to speed of transition. In turn, this will improve understanding of the relationship between transition speed and later outcomes and the stages of drug use.

Additionally, if a relationship is observed between this specific transition and later cannabis use outcomes this may provide a new marker for those who would benefit from intervention, regardless of whether or not they experience early onset of cannabis use. Cannabis initiation does not occur solely during adolescence, and research into US college students found that 25% of those sampled initiated

cannabis use after starting college, having not used it previously (Pinchevsky et al., 2012). If speed of the specific transition studied within this chapter is associated with later cannabis outcomes, after controlling for the effect of early initiation of cannabis use, it may prove useful as an early marker for intervention that is applicable to a wider population of cannabis users.

In this chapter I explore the relationship between the speed of this specific transition and later cannabis use, abuse and dependence outcomes, and identify the extent to which variance in the speed of transition to subsequent use of cannabis is attributable to genetic and environmental factors.

5.2 Aims and Hypotheses

Aim 1

Test whether speed of transition from initiation to subsequent use of cannabis is associated with increased likelihood of later daily cannabis use, abuse and/or dependence, and cannabis related treatment seeking when accounting for the influence of socio-demographic, childhood, mental health, peer and drug use factors that may confound the association

Hypothesis 1

Individuals who have faster transition from initiation to subsequent use of cannabis will have an increased likelihood of daily cannabis use, abuse and/or dependence and cannabis related treatment seeking later in life.

Hypothesis 2

Associations between speed of transition from initiation to subsequent use of cannabis and lifetime risks of daily cannabis use, abuse and/or dependence and cannabis related treatment seeking will persist after control for socio-demographic, childhood, mental health, peer and drug use factors identified as potentially confounding variables.

Hypothesis 3

Speed of transition from first to subsequent use of cannabis will be associated with later cannabis outcomes after controlling for early cannabis initiation.

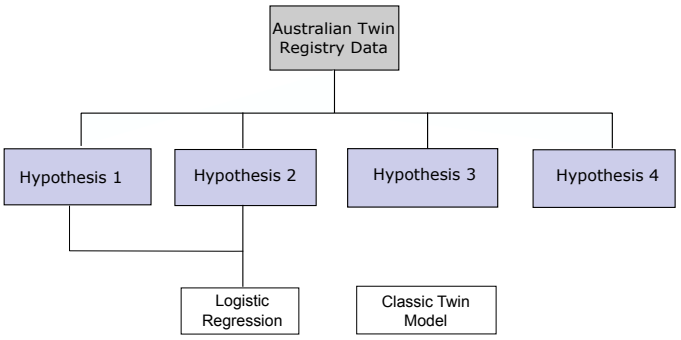
Aim 2

Examine the extent to which the speed of the transition is attributable to additive genetic, shared environmental or non-shared environmental effects.

Hypothesis 4
The speed of transition will be influenced by both additive genetic and environmental factors.

See Figure 5.1 for an outline of how the hypotheses of this chapter are addressed through two different analyses.

Figure 5.1: Dataset and analyses used to address the aims of this chapter



5.3 The Relationship Between Speed of Transition from Initiation to Subsequent Cannabis And Later Cannabis Outcomes

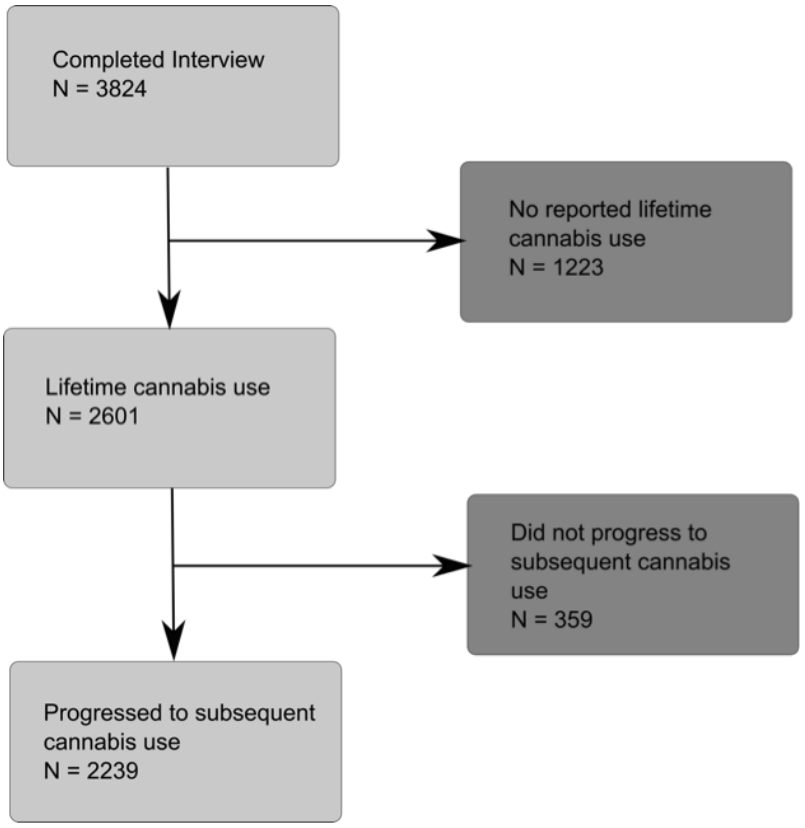
5.3.1 Methods

The first analyses in this chapter focus on the relationship between the speed of transition to subsequent use and later cannabis outcomes.

Sample

The sample for this portion of the chapter analyses was ATR participants who had progressed to subsequent cannabis use (used cannabis more than twice in their lives, see Figure 5.2).

Figure 5.2: Deriving the sample for the subsequent use analysis from the complete twin cohort



Independent and Dependent Variables

Speed of Transition to Subsequent Cannabis Use

Those who had reported using cannabis at least once in their lives were asked how long after their first use did their second use of cannabis occur. The categories in this analysis represent the underlying continuous construct of time to subsequent use, but continuous data were not available. These raw data were coded into 9 different categories, defined in terms of days, weeks, months or years. These original categories were collapsed into 4 discrete groups to overcome sparse cells: within 1 week (19.8%), within 3 months (but not including those who transitioned within 1 week) (37.7%), between 3 months – 12 months (21.7%), more than 1 year later (20.8%) (with those who had reported not trying cannabis for a second time excluded from the analysis, N = 359).

Cannabis Use Outcomes

The outcomes of daily cannabis use, abuse and/or dependence, and treatment seeking were used in the analyses (see Chapter 2 for variable derivation).

Analysis Plan

Analyses were conducted in Stata Statistical Software Version 11 (StataCorp, 2009). χ^2 tests and phi coefficients were used to assess the association between the speed of transition from initiation to subsequent use of cannabis and lifetime cannabis daily use, abuse and/or dependence and treatment seeking. Associations were deemed significant at the P $\leq$0.05 level. Logistic regression analysis was used to explore the association between the speed of transition from initiation to subsequent use of cannabis and the outcomes daily cannabis use, abuse/dependence, and treatment-seeking for cannabis use problems after adjustment for sociodemographic, childhood, mental health, peer and licit drug factors. An outline of the analysis is presented in Figure 5.3. To correct for the non-independence of observations (due to family clustering), Huber-White analysis for clustered data was implemented to provide robust standard errors. Post hoc comparisons across the varying speeds of transition were conducted using Wald χ^2 tests.

Identification of Covariates

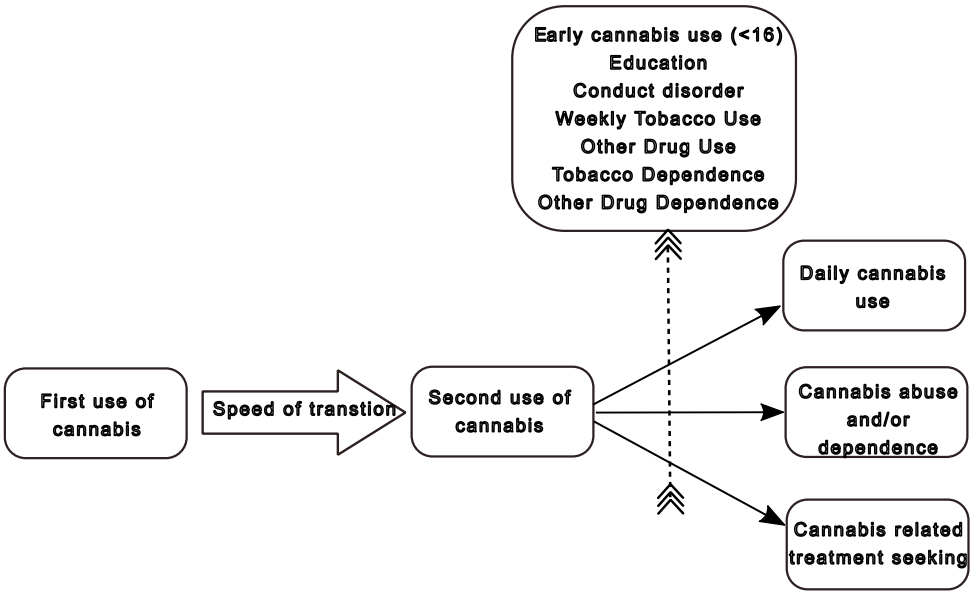
From the full list of potential covariates selected from the literature (see Chapter 2), those that may confound the association between speed of transition to subsequent cannabis use and later outcomes were identified through multivariate logistic regression. Covariates were included in the logistic models if significantly associated with both speed of transition (see Table 5.2) and the dependent variable (see Appendix 3).

5.3.2 Results

Analysis of the Relationship between Transition Speed and Cannabis Outcomes of Daily Use, Abuse and/or Dependence, and Treatment Seeking

In this sample 16.6% self-reported using cannabis daily during their period of heaviest use, 27.9% reported cannabis abuse and/or dependence, and 6%

Figure 5.3: Logistic regression model for the association between speed of transition to subsequent use and cannabis daily use, abuse and/or dependence, and treatment seeking

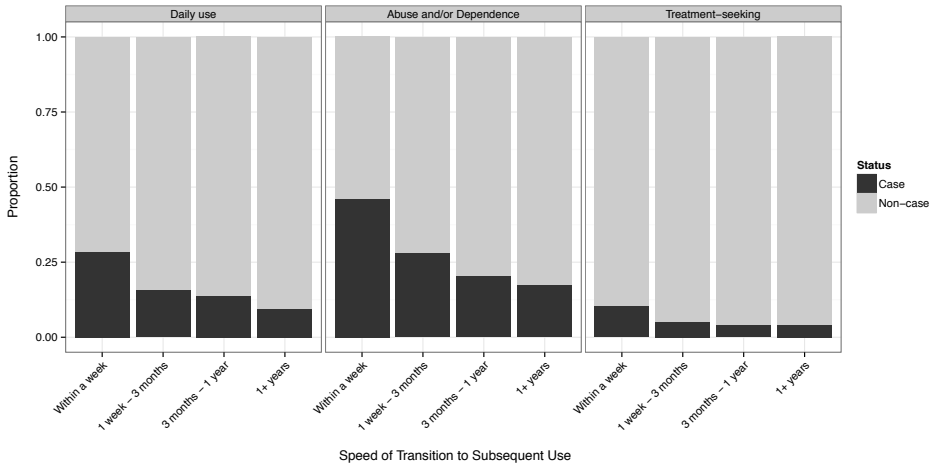


self-reported having discussed cannabis related problems with a professional. Proportions of each of the three cannabis use outcomes were significantly different between the transition speed groups (P $\leq$0.001 for all outcomes, see Table 5.1). For all outcomes the proportion that would go on to develop problems decreased approximately linearly across the groups (see Figure 5.4). Those whose second use of cannabis was within one week of initiation had the highest proportion of daily cannabis use (28.4%), abuse and/or dependence (46.0%), or cannabis-related treatment seeking (10.6%) (see Table 5.1).

Table 5.1: Association between speed of transition to subsequent cannabis use and daily cannabis use, cannabis abuse and/or dependence, and cannabis related treatment seeking in the ATR sample (N=2239)

Variable	Within a week N = 443	1 week - 3 months N = 844	3 months - 1 year N = 487	More than 1 year N = 465	Phi coefficient	χ^2 P value
	N (%)	N (%)	N (%)	N (%)		
Daily cannabis use N=372	126 (28.4)	134 (15.9)	67 (13.8)	45 (9.7)	0.17	$\leq$**0.001**
Cannabis abuse N=624 and/or dependence	204 (46.0)	238 (28.2)	100 (20.5)	82 (17.6)	0.22	$\leq$**0.001**
Treatment seeking N=132	47 (10.6)	45 (5.3)	21 (4.3)	19 (4.1)	0.10	$\leq$**0.001**

Figure 5.4: Diagram visualising the prevalence of cannabis outcomes by speed of transition to subsequent use



Association Between Speed of Transition to Subsequent Cannabis Use and Potential Model Covariates

After adjustment for other variables within the model, few factors were identified as significantly associated with the speed of transition to subsequent cannabis use analysis groups (see Table 5.2). These were level of completed education, CD, single parent family, lifetime weekly tobacco use and lifetime other drug dependence.

Adjusted Analysis of the Relationship Between Transition Speed and Later Cannabis Outcomes

After controlling for identified potentially confounding variables, those whose second use of cannabis was within a week were at increased odds of reporting daily use (OR 2.18, 95%CI 1.42-3.35) and meeting criteria for abuse/dependence (OR 2.73, 95%CI 1.92-3.90) (see Table 5.3). Those whose subsequent use of cannabis was within 3 months of initiation were at increased likelihood of reporting cannabis abuse and/or dependence (OR 1.53, 95%CI 1.10-2.11) (see Table 5.3). Post-hoc tests

identified that only the level 'within a week' was significantly different from both other levels, for the outcomes of cannabis daily use and abuse and/or dependence.

Speed of this transition was not significantly associated with cannabis treatment seeking; this association was non-significant regardless of whether early initiation of cannabis use was included in the model.

Table 5.2: Association between speed of transition to subsequent cannabis use and socio-demographic, childhood, mental health, peer and drug use factors in the ATR sample (N=2239)

Variable	Speed of transition to subsequent use			
	Within a week N = 443	1 week - 3 months N = 844	3 months - 1 year N = 487	More than 1 year N = 465
	N (%) OR (95% CI)	N (%) OR (95% CI)	N (%) OR (95% CI)	N (%) OR (95% CI)
Male gender	204 (46.1) 1.31 (0.96-1.77)	355 (42.1) 1.14 (0.88-1.47)	198 (40.7) 1.08 (0.82-1.44)	168 (36.1) 1.0
Lower level of completed education	159 (35.9) **1.78*** ** (1.30-2.44)	207 (24.5) 1.14 (0.86-1.51)	131 (26.9) 1.36 (0.99-1.85)	98 (21.1) 1.0
Conduct disorder	98 (22.1) **2.34*** ** (1.49-3.69)	103 (12.3) 1.48 (0.95-2.30)	45 (9.3) 1.32 (0.80-2.15)	35 (7.5) 1.0
Non-clinical depressive episode	2233 (52.7) 0.87 (0.65-1.17)	401 (47.8) 0.88 (0.69-1.12)	234 (48.2) 0.95 (0.73-1.23)	238 (51.5) 1.0
Parental alcohol problems	147 (34.3) 1.10 (0.80-1.51)	225 (27.1) 0.87 (0.66-1.15)	118 (24.6) 0.79 (0.58-1.07)	137 (30.0) 1.0
Parental drug problems	30 (6.9) 1.30 (0.66-2.55)	36 (4.3) 0.95 (0.50-1.79)	21 (4.4) 1.31 (0.67-2.59)	19 (4.2) 1.0
Single parent family	28 (6.3) 1.45 (0.73-2.88)	63 (7.5) **2.12*** (1.15-3.92)	25 (5.1) 1.29 (0.64-2.61)	1 (4.9) 1.0
High parental conflict	185 (44.6) 0.95 (0.70-1.28)	321 (41.0) 0.92 (0.71-1.19)	176 (38.1) 0.82 (0.62-1.10)	202 (45.3) 1.0
Strict parenting	238 (53.9) 0.93 (0.70-1.24)	411 (48.7) 0.84 (0.67-1.07)	231 (47.5) 0.79 (0.60-1.04)	248 (53.5) 1.0
High levels of peer cannabis use	56 (12.6) 1.38 (0.86-2.20)	87 (10.3) 1.25 (0.82-1.89)	28 (5.7) 0.73 (0.43-1.24)	38 (8.2) 1.0
Childhood sexual abuse	56 (12.8) 1.04 (0.66-1.63)	78 (9.3) 0.80 (0.53-1.19)	38 (7.9) 0.75 (0.48-1.19)	50 (10.9) 1.0
Infrequent childhood religious attendance	190 (42.9) 0.91 (0.68-1.21)	376 (44.6) 1.00 (0.79-1.28)	218 (44.8) 1.06 (0.81-1.39)	205 (44.1) 1.0
Lifetime monthly alcohol use	432 (97.5) 1.25 (0.55-2.82)	827 (98.0) 1.60 (0.80-3.21)	472 (96.9) 1.07 (0.52-2.21)	450 (96.8) 1.0
Lifetime weekly tobacco use	299 (67.5) **1.75*** ** (1.22-2.51)	492 (58.3) **1.38*** (1.01-1.89)	263 (54.0) 1.22 (0.85-1.75)	229 (49.4) 1.0
Lifetime other drug use	316 (71.3) 1.26 (0.93-1.71)	535 (63.4) 1.00 (0.77-1.29)	259 (53.2) 0.69 (0.52-0.92)	277 (59.6) 1.0
Lifetime alcohol dependence	182 (41.4) 1.09 (0.81-1.47)	290 (34.4) 0.98 (0.76-1.27)	155 (31.8) 1.00 (0.75-1.35)	148 (31.8) 1.0
Lifetime tobacco dependence	201 (45.4) 0.90 (0.62-1.32)	169 (34.7) 0.95 (0.67-1.33)	317 (37.6) 1.00 (0.68-1.48)	148 (31.8) 1.0
Lifetime other drug dependence	60 (13.5) **2.65*** ** (1.43-4.91)	60 (7.1) 1.64 (0.90-2.99)	34 (7.0) **2.32** ** (1.19-4.52)	17 (3.7) 1.0
Early initiation of cannabis (≤16)	0.83 (0.61-1.13)	1.15 (0.89-1.49)	0.85 (0.64-1.13)	1.0

*P ≤0.05 **P≤0.01 ***P ≤0.001.

Results 1 Summary

The analyses identified a significant association between speed of transition to subsequent use of cannabis and the outcomes daily cannabis use and cannabis abuse and/or dependence, which remained after controlling for identified confounding variables and for the effect of early initiation of cannabis use.

Table 5.3: Odds ratios (95% Confidence intervals) for association between speed of transition to subsequent cannabis use, covariates and later cannabis outcomes from logistic regression analysis of the ATR sample (N=2239)

Speed of Transition to Subsequent Use	Daily Use N = 372 Odds Ratio (95% Confidence Interval)		Abuse and/or Dependence N = 624 Odds Ratio (95% Confidence Interval)		Treatment Seeking N=132 Odds Ratio (95% Confidence Interval)	
	Univariate model	Adjusted model	Univariate model	Adjusted model	Univariate model	Adjusted model
Within a week N = 443	**3.71*** ** (2.55 - 5.38)	**2.26*** **[1] (1.47-3.49)	**3.99*** ** (2.92 - 5.44)	**2.87*** **[1] (2.00-4.11)	**2.79*** ** (1.60 - 4.85)	1.62 (0.90-2.95)
Within 3 months N = 844	**1.76*** ** (1.23 - 2.52)	1.33 (0.89-1.99)	**1.83*** ** (1.37 - 2.46)	**1.50** ** (1.08-2.08)	1.32 (0.77 - 2.28)	0.98 (0.55-1.74)
3 months - 1 year N = 487	**1.49*** (1.00 - 2.21)	1.49 (0.96-2.32)	1.21 (0.87 - 1.67)	1.18 (0.82-1.70)	1.06 (0.58 - 1.94)	0.95 (0.51-1.79)
More than 1 year N = 465	1	1	1	1	1	1
Covariate						
Male gender	**1.57*** ** (1.19-2.06)		**1.93*** ** (1.53-2.42)		1.40 (0.95-2.07)	
Lower completed education level	**1.57*** ** (1.19-2.08)		**1.28*** (1.00-1.63)		-	
Conduct disorder	**2.08*** ** (1.50-2.89)		**1.97*** ** (1.45-2.69)		-	
Lifetime weekly tobacco use	**4.51*** ** (3.20-6.36)		**2.86*** ** (2.25-3.65)		**3.36*** ** (1.85-6.09)	
Lifetime other drug use	**6.24*** ** (4.07-9.57)		**4.17*** ** (3.16-5.49)		**3.75*** ** (1.95-7.20)	
Lifetime other drug dependence	**2.55*** ** (1.76-3.69)		**2.94*** ** (2.01-4.30)		**4.02*** ** (2.59-6.26)	
Early initiation of cannabis (≤16)	**1.67*** ** (1.26-2.20)		**1.91*** ** (1.52-2.41)		**1.63*** (1.10-2.42)	

*P≤0.05 **P≤0.01 ***P≤0.001
[1]Identified as significantly different to other levels in post-hoc tests
All models have applied the Huber-White adjustment for family clustering

5.4 The Extent to Which the Speed of Transition is Attributable to Additive Genetic, Shared Environmental and Non-Shared Environmental Effects

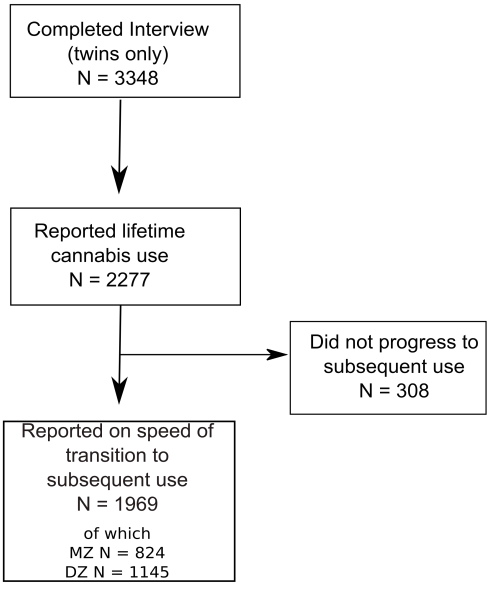
The next section of this chapter determines the extent to which the variance in speed of transition to subsequent use of cannabis is attributable to genetic and environmental factors.

5.4.1 Methods

Sample

The sample for this analysis is formed of the twins (siblings in sample excluded) who provided information on their progression to subsequent cannabis use (see Figure 5.5). The complete sample for the twin analysis included 824 MZ and 1145 DZ twins.

Figure 5.5: Derivation of the twin only sample for analysis of the speed of transition to subsequent cannabis use



Independent and Dependent Variables

Speed of Transition to Subsequent Cannabis Use

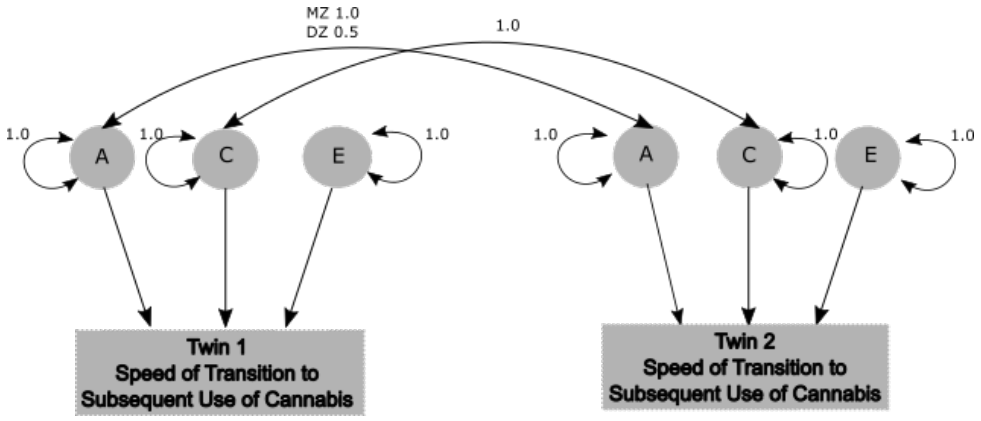
As outlined above, those who had reported using cannabis at least once in their lives were asked how long after their first use did their second use of cannabis occur, and continuous data were not available. The variable was coded as 4 discrete groups, and the twin-only group numbers were: within 1 week N = 400 (20.3%), within 3 months (but not including those who transitioned within 1 week) N = 746 (37.9%), between 3 months – 12 months N = 412 (20.9%), more than 1 year later N = 411 (20.9%) (with those who had reported not trying cannabis for a second time excluded from the analysis, N = 359). These figures are comparable to the group numbers for the complete sample (see outline above, Section 5.3.1).

Analysis Plan

Twin modelling was used to assess the extent to which variation in the speed of transition was attributable to additive genetic and environmental factors. Analyses were conducted using OpenMX (Boker et al., 2011) for the statistical software R (R Core Team, 2013). A full description of twin modelling methodology is provided in Chapter 2, section 2.5.5.

As there were low numbers of concordant twins, univariate analyses used raw ordinal data and full-information maximum-likelihood (FIML) estimation, which utilises data from twin pairs where data from a co-twin is unavailable. Model fitting was conducted using a step-wise approach. A liability threshold model estimating co-twin correlations was fitted to the data set and used to test assumptions regarding the equality of thresholds within and between MZ and DZ twin groups. Next, a univariate variance components model including adjustment for sex was fitted, partitioning the variance attributable to additive genetic (A), shared environmental (C), and unique environmental (E) factors (Figure 5.6). Difference in model fit was assessed via the likelihood-ratio χ^2 test and examination of the Akaike Information Criterion (AIC) (a description of these tests of model fit is provided in Chapter 2, Section 2.5.5).

Figure 5.6: Classic twin model to examine the extent to which speed of transition to subsequent use is attributable to additive genetic and environmental factors



5.4.2 Results

Additive Genetic and Environmental Factors Contributing to the Variance in Speed of Transition to Subsequent Cannabis Use

In order to find the most parsimonious twin model, birth order and zygosity were equated across groups, and the model fit compared against a saturated model whereby all parameters were freely estimated.

There was a 12.86 increase in -2LL from the saturated model to the model with equated parameters. This difference in fit was not found to be statistically significant (P = 0.17), and the model with equated parameters was accepted as the most parsimonious (see Table 5.4). For the model with equated thresholds, tetrachoric correlations were very similar between MZ and DZ twin pairs. Correlation was 0.25 for MZ twins, and 0.24 for DZ twins.

The estimate for additive genetic influences for the full model was small (0.002, 95%CI 5.80×10^{-8} – 0.35), and could be dropped from the model without a significant loss of fit (P = 1) (see Table 5.5). A model specifying only environmental influences (C and E) provided the best fit, with moderate shared environmental influences (0.25, 95%CI 0.15 – 0.34) and large unique environmental influences (0.75, 95%CI 0.66 – 0.84) on the variation in speed of this transition (see Table 5.5).

Table 5.4: Fit Indices and Tetrachoric Correlations for the Saturated Model and Equated Threshold Models of ATR Twins who Progressed to Subsequent Cannabis Use (N=1969)

	Parameters	Correlations MZ	Correlations DZ	-2LL	DF	AIC	Δ -2LL	Δ DF	P value
Saturated model	14	0.27	0.23	5263.22	1955	1353.22	-	-	-
Equate twin 1/twin 2 thresholds within MZ group	11	0.26	0.23	5266.79	1958	1350.79	3.57	3	0.31
Equate twin 1/twin 2 thresholds within DZ group	11	0.27	0.23	5267.76	1958	1351.76	4.54	3	0.21
Equate thresholds across MZ and DZ groups	5	0.25	0.24	5276.08	1964	1348.08	12.86	9	0.17

DF = degrees of freedom; AIC = Akaike Information Criterion; -2LL = -2 log-likelihood.

Table 5.5: ACE Model Fitting Results And Variance Components Point Estimates With 95% Confidence Intervals For Speed Of Transition From Initiation To Subsequent Use Of Cannabis in ATR Twins (N=1969)

Model	Proportion of Variance A (95% CI)	C (95% CI)	E (95% CI)	Model Fit Statistics -2LL	DF	AIC	Δ -2LL	Δ DF	P value
ACE Model	0.0002 (5.80×10^{-8} – 0.35)	0.25 (2.77×10^{-10} – 0.34)	0.75 (0.63 – 0.84)	5268.96	1963	1342.96	-	-	-
CE Model	- -	0.25 (0.15 – 0.34)	0.75 (0.66 – 0.85)	5268.96	1964	1340.96	0	1	1

DF = degrees of freedom; AIC = Akaike Information Criterion; -2LL = -2 log-likelihood.
Model adjusted for sex

Results 2 Summary

No additive genetic effect was observed to contribute to the variance in the speed of transition to subsequent use, with the majority of the variance attributable to unique environmental influence.

5.5 Discussion

5.5.1 Summary

A significant association was observed between speed of transition from initiation to subsequent use of cannabis and later likelihood of daily cannabis use and cannabis abuse and/or dependence. This association remained after controlling for potential confounders, and for early initiation of cannabis use. Those who experienced the fastest transition to subsequent use of cannabis were more than

twice as likely to progress to daily cannabis use and cannabis abuse and/or dependence.

The unique environment accounted for most (0.75) of the variance in the speed of transition from initiation of cannabis to subsequent use, and additive genetic effects could be dropped from the model without significantly affecting model fit. Given the absence of prior research on this transition these findings provide an original and intriguing contribution to the literature.

5.5.2 Relationship to Cannabis Outcomes

Previous research has found earlier initiation of cannabis use is associated with later problematic drug use/dependence (see Chapter 1), and by studying the novel transition from initiation to subsequent use this chapter has established that the association between speed of transition and later outcomes remains regardless of whether cannabis was initiated $\leq$16 years. This indicates that this transition may be useful to focus on for targeting intervention against problematic cannabis use, with the possibility of identifying a wider range of at-risk individuals than interventions focussing on early cannabis initiators.

5.5.3 Additive Genetic Contribution to Speed of Transition

Additive genetic effects had little observed influence on variation in the speed of this transition, which is in contrast to findings of moderate heritability for other transitions (Kendler et al., 1999a; Lynskey et al., 2012; Sartor et al., 2009a; Verweij et al., 2010). Speed of other specific transitions has been found to be moderately heritable, with 30% (95% CI 0.15–0.46) of the rate of transition from non-use to initiation attributed to additive genetic effects and similar findings observed for the rate of transition from initiation to first dependence symptom (36%, 95% CI 0.19–0.44) and first dependence symptom to the development of dependence (37%, 95% CI 0.00–0.58) (Sartor et al., 2008). In contrast, the present findings suggest the speed of transition from initiation to subsequent use of cannabis is predominately influenced by unique environmental factors.

The absence of observed variation attributable to additive genetic factors will require replication in future studies with a genetically informative design, as there

is potential for it to result from low power within the sample. Alternatively, the E parameter in twin modelling can represent measurement error (Plomin et al., 2013). As stated this is a novel transition measured through retrospective report, and other methods of measuring this transition would need to be applied in order to fully remove error as an explanation. However, a cautious interpretation of these results is that the speed of this transition acts as a proxy for an individual being within an environment in which drug access is easy (not measured within these data), and which facilitates progression to problematic cannabis use.

5.5.4 Limitations

Firstly, these data were based on retrospective self-report, which introduces the possibility of recall bias. Secondly, the measure of transition speed was comprised of relatively wide categories. Thirdly, there were a low number of twin pairs concordant for speed of transition from initiation to subsequent use, which was overcome through the use of raw data for the twin modelling. Ordinal analysis can result in lower power, and may result in an underestimate of the true liability correlation (Neale and Cardon, 1992). Fourthly, the study lacked temporal information on a number of covariates within the analysis, and including these variables in the analysis represents a cautious approach to adjustment for confounding variables, which may lead to under-estimation of the effect of this transition. Fifthly, whilst likely representative of base population (of Health and Welfare, 2014) the prevalence of lifetime cannabis use in this sample is relatively high which may limit generalizability.

5.5.5 Implications

The faster transition from initiation to subsequent use is unlikely to have a traditional causal relationship with cannabis dependence. The association likely reflects a combination of individual and contextual factors, such as availability, that surround the rapid escalation. If replicated in prospective research, these findings may have practical utility for clinical practice, with the prospect of translation into a clinically useful marker with which to identify individuals likely to benefit from intervention. These findings have also highlighted the utility of studying different

transitions in substance use to disentangle the complex aetiology of drug use disorders.

CHAPTER 6

Through the Looking Glass: Exploring Transitions in Heroin Use

6.1 Introduction

Throughout this thesis, analyses have focussed on transitions in cannabis use. What is unknown is the extent to which these findings will also apply to other drugs. Exploring similar hypotheses in a different drug class provides a test of whether the relationship between early transitions and later drug use outcomes may extend beyond cannabis use.

Heroin has been found to contribute the highest proportion of the illicit drug contribution to Disability Adjusted Life Years (DALYs) (Degenhardt et al., 2014), and consequently is a drug of global health concern. However, the base rate of heroin dependence in the general population is low, and in the twin sample only one participant reported opiate dependence. Consequently general population samples are not suitable for exploring this outcome, necessitating the use of samples selected for their heroin use.

Data from a purposively selected community sample and a treatment-seeking sample are analysed in this chapter. Given that those in treatment seeking populations will predominately be already experiencing dependence, the sample will lack variation in dependence status which precludes this outcome from

analysis. Therefore, outcomes explored in relation to heroin use will be dependence severity at treatment seeking, time to treatment seeking, overdose, heavy heroin use, and injecting behaviour.

In addition to testing for findings similar to those observed for cannabis use, a unique feature of heroin use within the UK is that it allows for exploration of an additional hypothesis: does route of administration influence speed of transitions between stages of drug use? Heroin can be delivered through multiple routes of administration, broadly split into injecting or non-injecting (Bridge, 2010). Injection introduces heroin straight into the bloodstream, provides a near instant effect and high bioavailability (Tas and Day, 2016) which makes it the most efficient route of drug administration. In contrast, chasing (inhaling vapour from heated heroin) and snorting (nasal administration) heroin produce slower absorption, have lower bioavailability and a decreased onset of effect (Tas and Day, 2016). Injecting increases the individual's risk of blood borne viruses (Lawrinson et al., 2008), dependence (Strang et al., 1998), and unintentional overdose (Martins et al., 2015). Indeed, overdose is the leading cause of mortality amongst heroin users (Degenhardt et al., 2011). Additionally, amongst those who have been injecting heroin for a long time there is the risk of injecting into the groin (found by Darke et al. (2001) to be implemented after 6-10 years of injecting heroin use), which is an injecting site associated with physical harms (Hillstrom et al., 1990; Maliphant and Scott, 2005).

This chapter utilizes two different data sets to study the effect of initial heroin administration routes of 1) injecting, 2) chasing and 3) snorting on speed of transition to daily heroin use, and the relationship between early transitions and the outcomes of dependence severity, time to treatment seeking, overdose, groin/neck injecting, and heavy heroin use. Prior to 1980, the predominant route of administration among London based heroin users was injecting, but after 1980 chasing became more prevalent as a method of administration (Strang et al., 1992). Thus at the time of the study the population sampled contained individuals who had initiated their heroin use by both major routes. By utilising a population with variation in the route of heroin administration, and a population who have entered treatment for heroin dependence, specific testing of the relationship between Initial Heroin Administration Route (IAR) and transition speed, and transition speed and later outcomes, is feasible.

6.2 Aims and Hypotheses

Aim 1

Examine whether there is a relationship between the speed of early transitions in heroin use and time to treatment seeking, dependence severity at treatment seeking, overdose, groin/neck injecting, and heavy heroin use.

Hypothesis 1

Those who experience faster transitions in their early use of heroin will have shorter time to treatment seeking, greater dependence severity at treatment seeking, increased likelihood of overdose, more likely to be groin/neck injecting and have experienced heavier heroin use than those who experience slower early transitions.

Hypothesis 2

The association between faster transitions in their early use of heroin and time to treatment seeking, dependence severity at treatment seeking, overdose, groin/neck injecting, and heavy heroin use will remain after adjustment for demographic variables.

Aim 2

Examine whether route of heroin administration is associated with speed of transition to daily heroin use.

Hypothesis 3

Those who initiate heroin use through injection will have a more rapid progression to daily heroin use.

Hypothesis 4

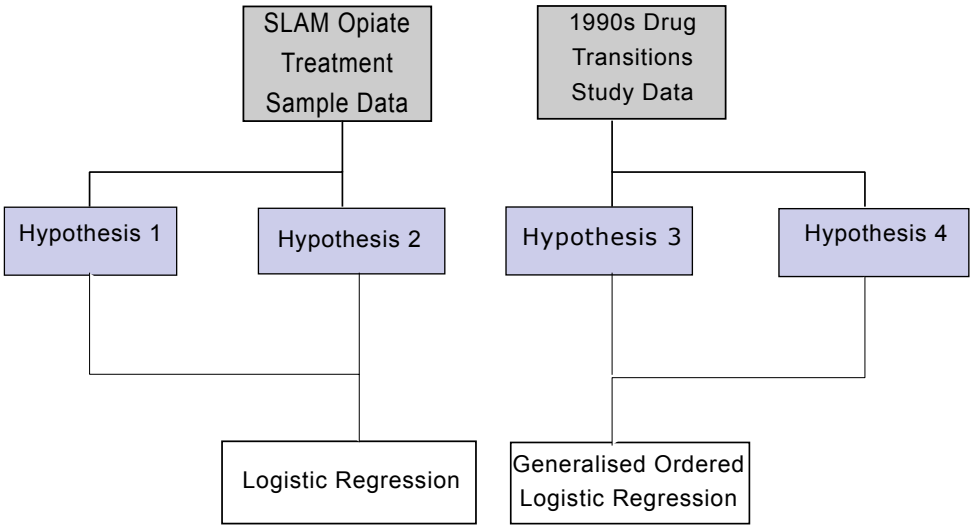
The association between injection of heroin and faster progression to daily heroin use will remain after adjustment for demographic variables.

See Figure 6.1 for an outline of how the hypotheses of this chapter are addressed through two different data sets.

6.3 Is Speed of Early Transitions in Heroin Use Associated with Later Heroin Use Outcomes

The first part of this chapter explores the association between speed of early transitions in heroin use and later outcomes associated with heroin use. Given

Figure 6.1: Dataset and analyses used to address the aims of this chapter (SLAM: South London and Maudsley NHS Trust)



that these analyses are conducted in a treatment-seeking sample, the outcomes (dependence severity at treatment seeking, time to treatment seeking, overdose, injecting behaviour and heavy heroin use) differ to those considered for cannabis in previous chapters.

6.3.1 Methods

Sample

This sample includes 93 participants who were in Opiate Substitution Treatment (OST) at the time of interview, recruited from South London and Maudsley Centres for Drug and Alcohol Treatment (see Chapter 2 Section 2.3 for full study description).

Independent and Dependent Variables

See Chapter 2, Section 2.3 for variable derivation. The transition speed variables included in the analysis were:

1. early opportunity to use heroin (early=$\leq$17);
2. time from opportunity to initiation of use;
3. time from initiation to second use of heroin.

See Chapter 2 Section 2.3 for full details of variable derivation.

The outcomes in the analysis were:

1. time to treatment seeking;
2. dependence severity at treatment entry (measured through the SDS);
3. overdose;
4. injecting into the groin/neck;
5. heavy heroin use.

As the outcomes of SDS score at treatment seeking and time from problem use to treatment seeking were not normally distributed (See Chapter 2 for measure definition and histogram), and would thus breach the assumptions of linear regression analyses, these potentially continuous outcomes were coded into binary variables and analysed accordingly.

To do so, (approximate) tertiles of the continuous variables were created. For SDS this provided a cut-off of a score of 14 and over for high SDS score (32.3%, N=30), and for time to treatment seeking those who sought treatment 7 or more years after the onset of their first problem were coded as having slow time to treatment seeking (31.8%, N = 28).

Analysis plan

Logistic Regression

Logistic regression was used to test whether transition speed was significantly associated with the later drug use outcomes. Univariate associations that were identified as significant ($P \leq 0.05$) were then entered into a multivariate logistic model.

Covariates in Regression Model

The regression model was adjusted for gender, age and ethnicity. See Chapter 2 Section 2.3 for a full outline of these covariates.

6.3.2 Results

Demographic Characteristics of the Sample

Most of the sample were male (74%), White British (56%), and reported both opiates and crack as their primary drug(s) used (59%) (see Table 6.1 for full characteristics).

Data on IAR was not collected in this sample.

Table 6.1: Characteristics of the SLAM sample (N=93)

Characteristic		N (%)
Gender	Male	69 (74)
	Female	23 (25)
Age	19 - 19	0
	20-29	5 (5.4)
	30 - 39	22 (23.7)
	40-49	51 (54.8)
	50 - 59	14 (15.1)
	60+	1 (1.1)
Ethnicity	White British	52 (56)
	Non-British White	21 (23)
	BME	20 (22)
Primary drug used	Opiates only	29 (31)
	Opiates and Crack Cocaine	55 (59)
	Other	9 (10)

Association between Speed of Early Transitions in Heroin Use and Later Time to Treatment-Seeking, Dependence Severity and Heroin Use Outcomes

Those who experienced early opportunity to use heroin (age 17 or under) were more than three times as likely to report overdose (OR 3.41, 95% CI 1.31 - 8.84) and injecting into the groin/neck (OR 3.83, 95% CI 1.53 - 9.57).

Significant associations were not observed between speed of transition from opportunity to initiation, or from initiation to subsequent use, and later heroin use outcomes. However, there was a moderately large association between speed of transition to subsequent heroin use and overdose (OR 2.10, 95% CI 0.92 - 4.82) and groin/neck injecting (OR 2.13, 95% CI 0.92 - 4.91). The lower confidence intervals on these results include 1.0, but only by a small amount. This may indicate that the current sample did not have sufficient statistical power to detect these effects (see Table 6.2). Power for this sample was calculated to detect an effect size of 3 or more (see Chapter 2 Section 2.3), and the effect size observed for the association between speed of transition to subsequent use was 2.10 for overdose and 2.13 for groin/neck injecting.

Table 6.2: Odds ratios (95% Confidence intervals) for association between speed of early transitions in heroin use and later heroin outcomes from logistic regression analysis (N=93)

Speed of Transitions	Outcomes				
	High SDS score at treatment seeking N=30 Odds Ratio (95% Confidence Interval)	Longer latency to treatment seeking N=28 Odds Ratio (95% Confidence Interval)	Overdose N=51 Odds Ratio (95% Confidence Interval)	Groin/Neck Injecting N=41 Odds Ratio (95% Confidence Interval)	Heavy heroin use N=32 Odds Ratio (95% Confidence Interval)
Early opportunity to use heroin N = 31	1.16 (0.46-2.89)	2.17 (0.84 - 5.63)	**3.41**** (1.31 - 8.84)	**3.83*** (1.53 - 9.57)	1.67 (0.66 - 4.23)
Opporunity to initiaiton: within a week N = 31	0.49 (0.20-1.25)	0.91 (0.35 - 2.36)	1.05 (0.44 - 2.54)	0.72 (0.30 - 1.74)	0.93 (0.37 - 2.37)
Initiation to subsequent use: wihtin a week N = 46	0.65 (0.27-1.57)	0.76 (0.31 - 1.86)	2.10 (0.92 - 4.82)	2.13 (0.92 - 4.91)	1.59 (0 .66- 3.82)

*P $\leq$0.05 **P$\leq$0.01 ***P $\leq$0.001.
All models have applied the Huber-White adjustment for clinic clustering

Multivariate Analysis of the Association between the Predictors Early Opportunity to Use Heroin and Early Initiation of Heroin Use and the Outcomes of Experiencing Overdose and Injecting into the Groin/Neck

Analyses were adjusted for the demographic variables of gender, age and ethnicity. After adjustment, those who reported early opportunity to use heroin were more than three times as likely to have experienced overdose (OR 3.28 95% CI 3.17 - 3.40) and injecting into the groin/neck (OR 3.82 95% CI 1.12 - 12.98) (see Table 6.3.2).

Table 6.3: Odds ratios (95% Confidence intervals) for multivariate analyses of relationship between early opportunity to use heroin and later overdose and groin/neck injecting in the SLAM sample (N=93)

	Outcomes	
Age of Opportunity to use Heroin	Overdose N=51 Odds Ratio (95% Confidence Interval)	Groin/Neck Injecting N=41 Odds Ratio (95% Confidence Interval)
	Adjusted model	Adjusted model
Early opportunity N=35	**3.28*** (3.17 - 3.40)	**3.82*** (1.12 - 12.98)
Later opportunity	1.0	1.0
Model covariates		
Male gender	**1.73*** (1.34 - 2.25)	**1.39*** (1.29 - 1.49)
Ethnicity		
White British	1.0	1.0
Other White	0.81 (0.64 - 1.03)	1.39 (0.44 - 4.35)
BME	0.42 (0.11 - 1.54)	0.66 (0.31 - 1.40)
Age at interview	0.99 (0.96 - 1.02)	0.97 (0.86 - 1.10)

*P ≤0.05 **P≤0.01 ***P ≤0.001.
All models have applied the Huber-White adjustment for clinic clustering

As converting continuous variables to categorical can be a source of bias in analyses (Altman, 1994), sensitivity analyses were conducted for early opportunity to use heroin, speed of transition from opportunity to initiation, and speed of transition to subsequent use of heroin. The analyses of different cut-off points indicated that results were not affected by the choice of age cut-off for transition speed variables (see Appendix 5).

Results Summary 1

Those who experience faster transition to heroin use opportunity are at increased likelihood of experiencing later overdose and groin/neck injecting than those who have later opportunity to use heroin. This association remained after adjustment for demographic variables. There was no association between speed of early transition and SDS score at treatment entry, time to treatment seeking, or heavy heroin use. There were no associations between the speed of transition from opportunity to initiation, or from initiation to subsequent use of heroin, and later heroin use outcomes; however, the results indicate this may result from a lack of power.

6.4 The Relationship Between Route of Administration and Speed of Transition

Route of heroin administration has been identified as a consideration in heroin transitions (Chapter 1). The analyses in this section of the chapter aim to examine the association between route of administration and speed of transition.

6.4.1 Methods

Sample

This sample was included 408 participants who had used heroin in the month prior to interview. Participants were recruited in the London area in 1991 (see Chapter 2 Section 2.4 for full study description).

Independent and Dependent Variables

The independent variable in these analyses was IAR. Dummy variables were created to for IAR, and these were used to test differences between IAR injecting and the other IARs, and between IAR chasing and the other IARs. The dependent variable was a categorical measure of time from initiation to daily heroin use. See Chapter 2, Section 2.4 for a full description of these variables.

Analysis plan

Initial tests of the association between IAR and speed of transition to daily heroin use were conducted using χ^2.

Testing the Suitability of an Ordered Logistic Model

The dependent variable of time to daily heroin use was a four level ordinal variable (1-3 weeks, 1 month – 11 months, 1-2 years, and 2+ years), which necessitated the use of an ordered logistic model. However, under the proportional odds assumption the relationship with the independent variable must be the same for each level of the dependent. This can be tested using likelihood ratio tests and the Brant test, with a significant result ($P \leq 0.05$) indicating assumptions are violated (Williams, 2006). In the present data, proportionality of odds assumption was violated for the dummy variable of IAR injecting (Likelihood ratio test P=0.02, Brant test P=0.01), the dummy variable of IAR chasing (Likelihood ratio test P=0.05, Brant test P=0.05), and year of heroin onset (Likelihood ratio test P=0.09, Brant test P=0.007).

The Generalised Ordered Logistic Model - An Alternative to the Ordered Logistic Model

When the proportional odds assumption is violated, a generalised ordered logistic model can be used (Williams, 2006). This allows variation in the relationship between the dependent variable and each level of the independent variable. This analysis produces multiple coefficients. The levels (L) of the dependent variable are analysed equivalent to a series of binary logistic regressions where the categories (C) of the dependent variable are combined. When C = 4 (as in the present analysis), for L = 1 category 1 is contrasted with categories 2, 3, and 4; for L = 2 the contrast is between categories 1 and 2 versus 3 and 4; and for L = 3, it is categories 1, 2, and 3 versus category 4. The levels of the dependent variable were 1 to 3 weeks, 1 month to within a year, 1 year, and more than 2 years. Coefficients represent the likelihood of those who inject heroin being in a more rapid transition group. Significance was determined at a level of $P \leq 0.05$.

Covariates in Regression Model

The regression model was adjusted for year of initiation (to account for cohort effects first reported by Strang et al. (1992), resulting from a rise in smoking of heroin in the UK around 1980), gender, ethnicity, regular drug use prior to heroin initiation, and current treatment status (to account for sampling effects). See Chapter 2 Section 2.4 for a full description of these covariates.

6.4.2 Results

Demographic Characteristics of the Sample

The majority of the sample were male (61.5%), white ethnicity (90.0%) and initiated their use of heroin through chasing (58.5%) (see Table 6.4). There were significant differences between IAR groups for year of initiation of heroin use ($P \leq 0.001$), with the majority of the sample who initiated heroin use after 1980 using chasing for their IAR (74.1% after 1980 compared to 23.1% of those initiating use before 1980). Prior to 1980, a greater proportion of the sample's IAR was through injecting (13.5% after 1980 compared to 57.0% of those initiating use before 1980) or snorting (12.4% after 1980 compared to 19.8% of those initiating use before 1980).

Table 6.4: Characteristics of the 1990s Drug Transitions Study sample (N=408)

Characteristic		N (%)
Gender	Male	246 (61.5)
	Female	154 (38.5)
Ethnicity	White	347 (90.1)
	African/Caribbean	26 (6.6)
	Asian	6 (1.6)
	Other	6 (1.6)
Initial route of Heroin administration (IAR)	Chasing	231 (58.5)
	Injecting	106 (26.8)
	Snorting	58 (14.7)
Time from initiation to daily heroin use	1 to 3 weeks	64 (18.1)
	1 month to 11 months	155 (43.9)
	1 -2 years	60 (17.0)
	2 or more years	74 (21.0)
Year of initiation	1979 or earlier	122 (30.5)
	1980 or later	278 (69.5)

Association Between IAR and Speed of Transitions to Daily Use

There were significant differences between the IAR groups in the number of participants who did not progress to daily use (see Table 6.5): while only 1 individual (1.0%) who initiated injecting did not progress to daily heroin use, 8.6% of those who initiated through snorting and 12.1% of those who initiated through chasing did not progress to daily heroin use. Of those who did progress to daily use, differences in speed of transition to daily use between IAR injecting and the other two IAR groups are shown in Figure 6.2.

Figure 6.2: Speed of Transition Amongst Those in the 1990s Drug Transition Study who Progressed to Daily Use (N=374), by Initial Administration Route

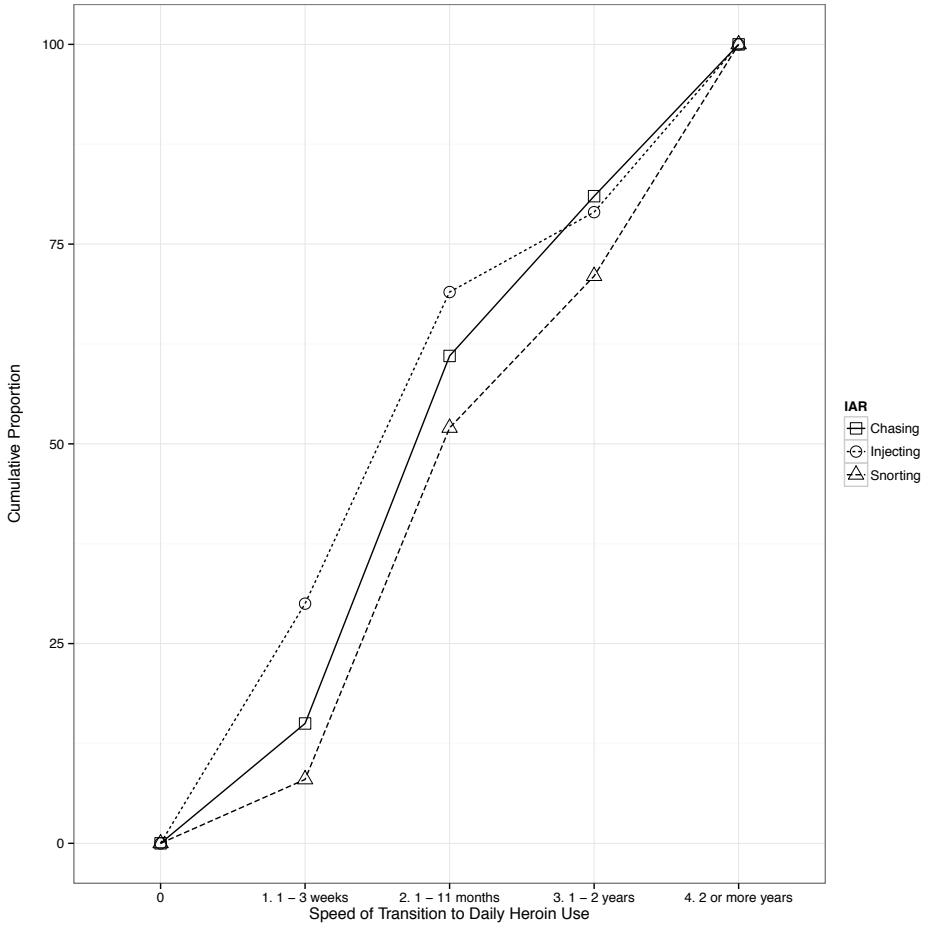


Table 6.5 demonstrates these differences were significant. Transition to daily use within 3 weeks of initiation was reported by 29.7% of those who initiated heroin use via injection, compared with only 14.8% of those who initiated via chasing and 7.7% of those who initiated via snorting.

Testing for proportionality of odds demonstrated that the assumption was violated for IAR and for year of heroin onset. The proportional odds assumption was relaxed for these variables using a generalised ordered logistic model.

Table 6.5: Association between route of initial administration and speed of transition to daily heroin use in the 1990s Drug Transition Study (N=408)

Progression to Daily Heroin Use	Initial Route of Heroin Administration (IAR) IAR injecting N = 106	IAR chasing N = 231	IAR snorting N = 58	
	N (%)	N (%)	N(%)	P value (χ^2)
Time to daily heroin use				
1 to 3 weeks N=63	30 (29.7)	29 (14.8)	4 (7.7)	**0.004**
1 month to 11 months N=153	40 (39.6)	90 (45.9)	23 (44.2)	
1 -2 years N=60	(9.9) 40	10 (20.4)	10 (19.2)	
2 or more years N=73	21 (20.8)	37 (18.9)	15 (28.9)	
Did not progress to daily heroin use	1 (1.0)	28 (12.1)	5 (8.6)	**0.003**

Multivariate Analysis of the Association between IAR and Speed of Transitions to Weekly and Daily Use

After adjustment, participants whose IAR was injecting were found to be more than four times as likely (OR 4.71, 95% CI 1.34 – 16.5) to progress to daily use within 1-3 weeks of initiation compared to other IARs (see Table 6.4.2). There were no significant differences in transition speed between the non-injecting IARs. None of the covariates remained significantly associated with speed of transition after adjustment.

Table 6.6: Univariate and Multivariate Odds ratios (95% CI) for the association between initial route of heroin administration (IAR) and speed of progression to daily heroin use in the 1990s Drug Transition Study (N=374)

IAR	Speed of Transition to Daily Heroin Use					
	1 to 3 weeks N = 63		1 month to 11 months N = 153		1 -2 years N - 60	
	Unadjusted model	Adjusted model	Unadjusted model	Adjusted model	Unadjusted model	Adjusted model
Injecting	**5.07****	**4.71***	2.09	1.01	1.54	1.00
	(1.68 – 15.3)	(1.34 – 16.5)	(1.05 – 4.16)	(0.49 – 2.08)	(0.72 – 3.33)	(0.43 – 2.32)
Chasing	2.08	1.93	1.43	0.89	1.74	1.00
	(0.70 – 6.22)	(0.55 – 6.72)	(0.77 – 2.65)	(0.46 – 1.73)	(0.87 – 3.50)	(0.46 – 2.17)
Model Covaraites						
Year of onset	1.01	1.00	0.98	1.00	0.94**	0.99
	(0.97 – 1.05)	(1.00 – 1.00)	(0.95 – 1.01)	(1.00 – 1.00)	(0.90 – 0.98)	(0.95 – 1.03)
		Unadjusted model			Adjusted model	
Ethnicity		1.45 (0.79 – 2.68)			1.00 (0.52 – 1.90)	
Regular drug use prior to heroin initiation		0.76 (0.50 – 1.14)			0.73 (0.47 – 1.13)	
Gender		1.26 (0.85 – 1.87)			1.00 (0.66 – 1.52)	
In treatment at time of interview		1.20 (0.81 – 1.77)			1.00 (0.64 – 1.57)	

*P ≤0.05 **P≤0.01 ***P ≤0.001.

Results Summary 2

Initiation of heroin use through injection was associated with a more rapid progression to daily heroin use; an association that remained after adjustment for demographic variables.

6.5 Discussion

These analyses confirmed that, to some extent, the relationship between speed of transition and later drug use outcomes that was observed for cannabis also applies for heroin. Those who reported early opportunity to use heroin were more than three times as likely to experience overdose and to report injecting into the groin/neck. Additionally, analyses in this chapter have identified that IAR is associated with speed of transition in heroin use. Participants whose IAR was through injection (compared to those who initiated through chasing or snorting) were more than four times as likely to use daily within a month of initiation.

6.5.1 Speed of Early Transitions in Heroin Use and Later Heroin Use Outcomes

Contrary to hypotheses, there was no association between early transitions in heroin use and dependence severity, time to treatment seeking, or heavy heroin use. Findings amongst alcohol dependent populations have previously identified associations between younger age of dependence onset and increased severity of alcohol problems (measured in number of symptoms, and length of dependence episodes), as well as longer time to treatment (Hingson et al., 2006). The null findings in the present thesis have plausible explanations. For dependence severity, there may be issues with the measure used. The SDS has not been validated as a retrospective measure, but was applied retrospectively in the thesis data collection in order to assess dependence severity at treatment entry. The findings may also result from low variation in speed of early transitions amongst the treatment population. For the measure of speed of transition to subsequent use, over 50% of the population reported a very rapid transition of within a week. This is in contrast to the observations for cannabis use in the general population ATR sample,

whereby only 19.8% reported such a rapid transition (see Chapter 5, Section 5.3). The reduced variation in speed of early transitions in this sample may reflect previous findings that transitions are faster in heroin use compared to other drugs (O'Keefe et al., 2016), or may result from the use of a population solely comprised of participants who have experienced problem use. Alternatively, it may be that severity of dependence is simply not associated with early transitions, and is a function of factors such as duration of heroin use (Barrio et al., 2001).

Earlier opportunity to use heroin was associated with increased likelihood of both overdose and injecting into the groin/neck. This is similar to findings in the literature that younger age of heroin initiation is associated with overdose (Lynskey and Hall, 1998); an association that remained after controlling for duration of heroin use, which was not possible in the present analyses (despite participants being OST it is not certain whether or not they were using heroin at the time of interview). The relationship between early opportunity to use heroin and increased likelihood of groin/neck injecting has not previously been observed.

Given the cross-sectional nature of the data no conclusions about causality can be drawn. However, it is plausible that those who have earlier opportunity to use heroin are exposed to environmental risk factors that increase their likelihood of later experiencing overdose. The heroin using population is one that is already at risk of a range of harms, with overdose the leading cause of mortality (Degenhardt et al., 2011). These findings relating to age of opportunity have potential clinical relevance for identifying, and providing harm reduction interventions to, groups who may be at especially high risk of overdose and chronic heroin use.

6.5.2 The Relationship Between Route of Administration and Speed of Transition

Participants who initiated their heroin use through injecting progressed much more quickly to daily heroin use than those using other methods. Injection of similar doses produces a high concentration of the drug in the bloodstream (Rang and Dale, 2016), and has higher bioavailability than smoking administration (Tas and Day, 2016). Injection is already known to be associated with greater likelihood of progressing to dependence as a result of the increased efficiency of drug delivery (Strang et al., 1998), and it is plausible that a similar mechanism is underlying the

faster transition to daily use amongst participants whose IAR is through injection.

Non-injecting administration has previously been shown to minimise harms, reducing likelihood of developing dependence (Strang et al., 1998), overdosing (Tas and Day, 2016) and contracting blood-borne viruses (Strang et al., 1998). The increased latency to daily use amongst participants whose IAR is chasing or snorting findings support the harm-reduction case for encouraging non-injecting routes of administration (Bridge, 2010; Hunt et al., 1998; Pizzey and Hunt, 2008; Strang, 1988).

6.5.3 Limitations

There are limitations of this study that must be considered when interpreting the results. Data were collected on only a small number of potential covariates, and consequently detailed analysis of what may underlie the relationship between age of opportunity to use heroin and overdose cannot be conducted. The data were collected through self-report, which is unlikely to bias results in samples of injecting drug users (Darke, 1998). However, the use of retrospective self-report introduces the potential for recall bias to affect the results. The present findings would need to be replicated in prospective research. Additionally, these findings would benefit from replication within a new cohort to ensure that the results are not an artefact of the drug market at the time of data collection (Horyniak et al., 2015).

6.5.4 Implications

The findings support the case for encouraging non-injection as a route of administration (Bridge, 2010; Wodak and McLeod, 2008) as it was associated with slower progression to daily injecting. Further, the faster transition to daily use through injection has implications for drug treatment provision. The need for drug services to attract those whose treatment needs are less immediate, in order to encourage those already injecting to switch to non-injecting, has previously been suggested in the literature (Strang, 1988). Focus may benefit from shifting to target those who have recently begun injecting, to provide encouragement to non-injecting administration (or indeed, heroin abstinence) (Pizzey and Hunt, 2008). However, intervening at the treatment service level could never be expected to reach the

complete population of individuals who have recently initiated heroin use; this requires changes at the policy level. One idea that has previously been suggested is that of altering policing to penalise the supply of injectable heroin whilst being more tolerant of the supply of heroin that could only be smoked (Strang and King, 1997). The findings of this study suggest that doing so has the potential to limit the harms of heroin use and reduce the prevalence of dependence by not only reducing the number of people who use heroin who progress to daily use, but also to allow a greater window for treatment services to reach people who use heroin before a daily habit has developed.

CHAPTER 7

Discussion

7.1 Summary of Findings

The analyses presented in this thesis have demonstrated that addiction is a multi-stage process, and that studying transitions in drug use contributes novel insights into the aetiology and developmental course of SUDs. Consistency has been identified in not only the individual and external factors associated with multiple stages of drug use, but also the additive genetic influences acting on stage progression. Additionally, speed of early transitions has emerged as a potential marker of later problematic drug use. Taken together, these results provide answers to a number of important questions relating to the course of SUDs.

7.1.1 Is Transition Speed Associated with Later Likelihood of SUDs?

I explored the relationship between the speed of early cannabis transitions and later cannabis daily use, abuse and/or dependence, and treatment using a general population sample. Age of onset of opportunity to use cannabis at age 14 (relative to those who had opportunity age 18 or over) was associated with more than twice the likelihood of cannabis daily use, abuse and/or dependence, and treatment seeking. Age of onset of opportunity to use cannabis at age 15-16 (relative to those who had opportunity age 18 or over) was associated with almost

twice the likelihood of daily cannabis use, and twice the likelihood of cannabis abuse and/or dependence. Age of opportunity age 17 (relative to those who had opportunity age 18 or over) was associated with increased likelihood of cannabis abuse and/or dependence. Transition to subsequent use within a week of cannabis initiation (relative to those whose transition was more than a year after initiation) was associated with twice the likelihood of cannabis daily use and abuse and/or dependence. Transition to subsequent use within three months of cannabis initiation (relative to those whose transition was more than a year after initiation) was associated with increased likelihood of cannabis abuse and/or dependence.

7.1.2 What is the Profile of Individual, Childhood, Mental Health And Other Drug Use Factors Associated with Speed of Transition to Opportunity to Use and Dependence?

I investigated factors that influence the speed of early transitions, and whether the influence of these factors is consistent across transition stages. Again, these analyses used a general population sample for which data on individual, childhood, mental health and other drug use factors were available. I found that although some factors were consistently associated with the speed of transition through different stages of drug use, others displayed unique associations with specific transitions. Conduct Disorder (CD), parental drug problems, weekly tobacco use (preceding onset of stage) and male gender were consistently associated with faster progression to opportunity to use cannabis, and from opportunity to dependence. The results demonstrated that the unique influences on speed of transitions are proximal factors, such as

1. parental conflict, childhood religious attendance, Childhood Sexual Abuse (CSA) and parental alcohol problems increasing the rate of progression to opportunity;

2. other drug and depressive disorders increasing rate of progression to cannabis dependence.

7.1.3 Are Genetic Influences on Speed of Early Transitions Correlated with Genetic Influences on Dependence?

Leveraging the genetically-informative nature of the general population sample used in previous analyses, I conducted a classical twin study in order to examine the extent to which the speed of early stage transitions in cannabis use are influenced by additive genetic and shared environmental influences. This also allowed me to explore the extent to which genetic and environmental influences on these transitions are unique or are correlated with those influencing liability to cannabis dependence. The study identified that additive genetic effects accounted for the majority of the variance in the age of opportunity to use cannabis (66%) and cannabis abuse and/or dependence (78%), and that these overlapped with the genetic effects acting on the development of cannabis abuse and/or dependence (42%). However, there was no evidence that genetic factors influenced liability to the novel specific transition from initiation to subsequent cannabis use.

7.1.4 Is Speed of Transitions in Heroin Use Associated with Outcomes Beyond Dependence, and Route of Administration?

In the final component of this thesis, I investigated drug transitions in a clinical, rather than general population, sample. I explored the relationship between the speed of early heroin transitions and later heroin dependence severity, overdose, injecting behaviours and heavy heroin use. Given that injecting administration of heroin is associated with worse health outcomes than non-injecting administration (Strang et al., 1998), the effect of Initial Heroin Administration Route (IAR) on speed of transition was hypothesised to have a differential effect on speed of transition to daily heroin use. I established that those who reported an earlier opportunity to use heroin were more than three times as likely to experience overdose and groin and/or neck injecting, and that those who initiated their heroin use through injecting, rather than snorting or chasing heroin, had a four-fold increase in speed of progression to daily heroin use.

7.2 The Value of Considering the Multi-Stage Nature of Drug Use

As outlined in Chapter 1, the development of SUDs is a multi-stage process in which specific transitions must occur. In this thesis I focussed on the early drug use stages of opportunity to use and progression from initiation to subsequent use in cannabis and heroin use. This included the transition from opportunity to dependence for cannabis only, and the transition from initiation to daily use for heroin only.

Studies of dependence commonly compare individuals who are drug dependent against those who are not dependent, which leads to uncertainty about the stages in the development of drug dependence at which specific genetic or environmental influences are most prominent (Nelson et al., 2013). By applying a multi-stage approach in this thesis, estimates of influences on progression through the stages of drug use were not conflated. Consequently, there were a number of unique findings that contribute to our understanding of SUDs and their aetiology.

Evidence from studies of tobacco and alcohol use has previously suggested that, in their aggregate, genetic influences are relatively weaker at earlier stages in the development of drug dependence (see Lynskey et al. (2010) for review). In the analyses presented in this thesis, the estimated heritability of age of opportunity to use cannabis was 0.66, and the estimated heritability of cannabis abuse and/or dependence was 0.78. This represents a strong contribution from additive genetic effects to the variance in both opportunity to use and abuse and/or dependence, and whilst the estimate for opportunity is smaller, the difference is not large. However, my analyses of subsequent use of cannabis did not provide any evidence for an additive genetic contribution to the variance of speed of this transition. The results of this thesis have demonstrated variation in the extent to which genetic effects influence the early stages of drug use.

Considering opportunity to use cannabis and the development of cannabis abuse and/or dependence as distinct stages of drug use allowed exploration of the extent to which they were influenced by shared genetic effects. Of these genetic effects, 42% were shared, and 46% of genetic effects on cannabis abuse and/or dependence are unique from those acting on opportunity. These (potentially surprising) findings will be discussed in greater detail later in this chapter. It

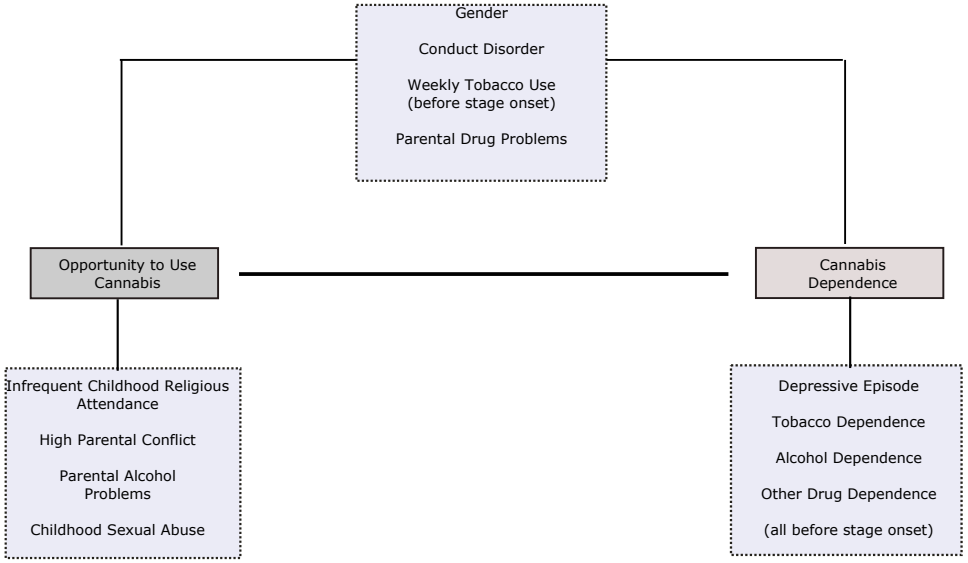
has been speculated that early stages of drug use may be genetically influenced through personality traits such as novelty seeking (Laucht et al., 2007), which is an example one of the many factors that may account for the overlap in genetic influences. With regards to the factors uniquely associated with dependence, genetic influences on drug metabolism may be more likely to exert influence at this stage (Dick et al., 2014). Similar findings from tobacco research, using genetic risk scores created using multiple single-nucleotide polymorphisms (SNPs) identified as associated with number of cigarettes smoked per day, has identified that this risk was unrelated to initiation of tobacco use but that higher genetic risk score was significantly associated with increased risks for the development of nicotine dependence (amongst other progression outcomes) (Belsky et al., 2013). Consequently the present findings support research indicating that at least some of the genetic influences on drug use differ at different stages, and suggest that research identifying the unique gene associations is warranted.

I identified distal and proximal risk factors that were unique to specific stages of drug involvement, while others acted across multiple stages. CD, weekly tobacco use, parental drug problems and gender were found to influence the speed of studied transitions in cannabis use. However, a number of unique factor associations were observed for each transition; these were predominately proximal influences. Previous research into stages of alcohol use has demonstrated differences in association by stage of use for environmental factors (Sartor et al., 2007). Interestingly, a number of the factors identified as unique to alcohol dependence (nicotine dependence, cannabis abuse, generalised anxiety disorder) broadly reflect those found to be specific to progression to dependence in the present analyses.

7.3 What Factors Act Across Stages of Drug Use?

One advantage of the approach taken in this thesis is that a consistent selection of factors was tested for their relationship to progression through different stages of drug use. Figure 7.1 provides a visualisation of these results for clarity of understanding. The following factors were identified as being associated with transition across drug use stages; these are predominately distal factors; influences that are further removed from the onset of behaviour, as opposed to proximal

Figure 7.1: Factors associated with progression through the stages of drug use



factors which act directly on the onset of that behaviour.

7.3.1 Conduct Disorder

CD was associated with faster progression to cannabis use opportunity, and from opportunity to dependence. This finding is in line with existing research linking CD in adolescence to both cannabis use and problematic cannabis use (Fergusson et al., 1993; Heron et al., 2013; Pedersen et al., 2001; Zohsel et al., 2016). Additionally, CD has been linked to speed of transition to cannabis initiation (Galéra et al., 2010) and from initiation to problem use (Sartor et al., 2013a), with childhood aggressive or disruptive behaviour associated with earlier opportunity to use cannabis in males' (Storr et al., 2011). The analyses in this thesis add to this literature, identifying further transitions that CD is associated with.

The effect sizes for the association with CD differed for each transition, with an estimated adjusted HR of 5.57 (95% CI 1.52-20.47) for age of opportunity and HR 2.49 (95% CI 1.91-3.25) for transition from opportunity to dependence. It is important to note that the confidence intervals indicate that effect sizes may overlap. However, the difference in effect sizes across transitions does mirror the trajectories of conduct problems. Conduct problems have been shown to split into four classes: those limited to childhood, those that have early onset

and persist into adolescence, those that have adolescent onset, and no problems (Barker and Maughan, 2009; Moffitt, 1993). These trajectories have been shown to be differentially associated with cannabis use and problem use, with those in the adolescent onset class at increased likelihood of cannabis use (OR 1.89, 95% CI 1.24 - 2.88) and those in the early-onset persistent class at increased likelihood of problem cannabis use (OR 4.97, 95% CI 2.94 - 8.41) compared to a low problem group (Heron et al., 2013). Similarly, research splitting CD into different dimensions and exploring trajectories within those identified classes has identified associations with substance use (Reef et al., 2010). Within the CD dimension of Status Violations, which includes drug and alcohol use, a group was identified who exhibited increased engagement in these behaviours between the ages 4-18. This group was at increased likelihood of substance abuse/dependence compared to those who had low engagement in the behaviours (OR 11.7, 95% CI 3.4–40.2). Those who only had a medium increase in these behaviours between the ages 4-18 also had an increase in likelihood of substance abuse/dependence, but to a lesser extent than those with high increase (2.3, 95% CI 1.4–3.8). It is likely that differences in the class of CD will be related to speed of transition in cannabis use, and may underlie the changes in effect size over time.

The association between speed of transition in cannabis use and CD may have a genetic basis. The genetic influences associated with CD have been found to correlate with the genes associated with cannabis use (r=0.28 in males, r=0.09 in females) (Verweij et al., 2016). In a separate study, the genetic correlation between CD and cannabis abuse/dependence was estimated at r=0.72 (Grant et al., 2015). Additionally, the genetic overlap between alcohol, tobacco and cannabis use disorders was attributable to genes shared with CD. In the present study, age of opportunity to use cannabis and cannabis abuse and/or dependence were found to have correlated genetic influences (42%). It is plausible that at least some of these shared genetic influences may overlap with those acting on CD.

7.3.2 Weekly Tobacco Use

Weekly tobacco use (preceding cannabis use stage onset) was associated with faster progression to cannabis use opportunity, and from opportunity to dependence. This is in line with existing research that identified that those who

reported lifetime use of at least 100 cigarettes had an earlier opportunity to use cannabis (Agrawal et al., 2013), and that those who were regularly smoking tobacco at least once a week for 2 months prior to problematic cannabis use (and had smoked greater than 20 cigarettes in their lifetime) had faster progression from initiation to their first symptom of problematic use (Sartor et al., 2013a).

This finding is likely partially attributable to the shared typical route of tobacco and cannabis administration (smoking). It may be expected that individuals who are regularly smoking tobacco may have faster transitions in cannabis use as a result of familiarity with the administration method. Additionally, tobacco and cannabis are often administered simultaneously (Agrawal et al., 2012), which may contribute to a relationship between tobacco use and speed of cannabis transitions. Recent research has found that, globally, it is more common to administer cannabis with tobacco than without, and in Australia (where the analysis sample was drawn from) 51.6% of those surveyed were likely to use cannabis concurrent with tobacco (Hindocha et al., 2016). Additionally, research suggests that common genetic influences act on initiation of tobacco use and the initiation of illicit drugs, including cannabis (Huizink et al., 2010). Thus shared administration route, concurrent use, and shared genetic factors may, in combination, explain the finding that regular tobacco use is linked to transitions across the stages of cannabis use.

7.3.3 Parental Drug Problems

Participant report of one or both parents experiencing problems with any drug was associated with younger age of opportunity to use cannabis, and with faster progression from opportunity to the development of cannabis dependence. These findings are in line with existing research suggesting that parent substance problems increase likelihood of drug use opportunity, but this existing research did not find association between parent substance problems and SUD (Benjet et al., 2013); this may result from a lower prevalence of SUDs in the population in which this previous research was based. Associations have also previously been found between parent illicit drug use (not necessarily problematic) and higher frequency of offspring cannabis use at age 15-16 (Fergusson and Horwood, 1997) or vulnerability to substance use (Lynskey et al., 1998), and familial transmission has been identified for SUDs in studies of the families of those in treatment for

SUDs (Merikangas KR et al., 1998; Miles et al., 1998). That these factors are also associated with speed of progression to SUDs is a novel finding.

There are a number of potential mechanisms through which parent drug problems could influence progression through the stages of drug use. One is genetics, which will be discussed in detail later in this chapter (Section 7.5). A second is family attitudes towards drug use, which have been highlighted in a comprehensive review of the literature as a risk factor for SUDs (Hawkins et al., 1992). However, regardless of attitudes towards their child's drug use, parent engagement in behaviour alone may have an effect. In studies of alcohol use, parent alcohol use has been found to increase likelihood of adolescent alcohol use even when the parent is opposed to adolescent drinking (Hung et al., 2015). Although more difficult to test in drug use due to the reduced prevalence of the behaviour compared to alcohol, the relationship between parental drug problems, family attitudes towards drug use, and the progression through the drug use stages may prove to be a fruitful avenue for further exploration.

Another mechanism through which parental drug problems may lead to earlier opportunities to use cannabis, and faster progression to development of dependence, is the effect of parent drug use on a child's drug access. It has previously been found that cannabis availability accounted for almost all (97%) of the shared environmental (childhood and family environment) effects acting on cannabis initiation and developing symptoms of cannabis abuse (Gillespie et al., 2009). This may reflect direct availability, or parenting practices that are conducive to drug exposure. Applied to the results of this thesis, it is plausible that one mechanism through which progression to opportunity and dependence is increased is through individuals whose parents experienced drug problems growing up in environments that increased cannabis availability.

7.3.4 Gender

In the analyses presented in this thesis, gender was associated with earlier age of opportunity to use cannabis and faster progression from opportunity to dependence, with males at increased likelihood of making faster transitions. This finding is in contrast to the majority of the existing research in this area. Differences have not previously been found between males and females for age of opportunity

to use cannabis (van Etten and Anthony, 1999), for progression from initiation within a year of opportunity to use cannabis (van Etten et al., 1999), or for speed of transition from initiation of cannabis to problem use (Haas and Peters, 2000; Ridenour et al., 2006). Gender differences in drug use and the development of SUDs have previously been observed (Young et al., 2002), although there is a suggestion that these differences are less pronounced in those aged 18-29 compared to those in older cohorts (Degenhardt et al., 2008), which may indicate that drug use is becoming more equal in younger generations.

Sex differences in *lifetime* drug use opportunity (not studied in the present thesis) have previously been found. It has been observed that males are more likely to have the opportunity to use drugs (Caris et al., 2009; van Etten et al., 1999), but are no more likely than females to use that drug once the opportunity has occurred (van Etten et al., 1999). It has been speculated that these differences may result from males being involved in activities associated with higher drug exposure risk, or that they may receive lower parental monitoring (van Etten and Anthony, 1999). These may contribute to gender differences in speed of transition to opportunity and dependence.

7.4 Which Factors Drive Progression to Individual Stages of Drug Use?

The factors identified as uniquely associated with progression through specific stages of cannabis use can be described as proximal factors. An overview of the factors associated with progression to specific stages of drug use is provided in Figure 7.1.

Factors uniquely associated with earlier age of opportunity to use cannabis were infrequent childhood religious attendance, high parental conflict, parental alcohol problems, and CSA. None of these factors have previously been found to be associated with younger age of opportunity to use cannabis, but childhood religious practices are associated with decreased likelihood of cannabis use opportunity (Chen et al., 2004), as are the parenting factors of lower parental involvement and higher levels of coercive discipline (Chen et al., 2005b). It is perhaps surprising that CSA is uniquely associated with this stage given that the literature review (Chapter

1, Section 1.5) identified this factor was associated with speed of progression to cannabis use disorder (Sartor et al., 2013a, 2015).

The factors uniquely associated with speed of transition from opportunity to use cannabis to dependence indicate that a pattern of co-occurring substance use and dependencies develops between the onsets of these stages. One explanation for the development of co-occurring dependencies is that the same factors are underlying dependence on different substances (Agrawal et al., 2004; Morral et al., 2002). Research using twin populations has found that the genetic and shared environmental factors underlying use and abuse/dependence for cannabis, cocaine, hallucinogens, sedatives, stimulants, opiates, inhalants, and prescription drugs were not drug-specific (Kendler et al., 2003; Tsuang et al., 1998), although other research has found more evidence for drug-specific genetic factors acting on the development of dependence (Palmer et al., 2012; Sartor et al., 2010). The finding that preceding dependence on other substances is associated with faster progression to cannabis dependence supports the literature suggesting some factors increase liability to multiple dependencies.

Non-clinical depressive episode onset (preceding the development of cannabis dependence) was associated with faster progression to cannabis dependence. Research into the relationship between cannabis dependence and depression has not established a consistent causal role for cannabis use in the development of major depressive disorder (Danielsson et al., 2016; Feingold et al., 2014; Gage et al., 2015). Research on the association in the other direction, as tested in this thesis, has found association between major depressive disorder and incident cannabis abuse, but not dependence (Pacek et al., 2013). The self-medication hypothesis is a plausible explanation for the association observed within this thesis. Those in treatment for drug abuse have been found to report using drugs to relieve symptoms of depression (Weiss et al., 1992), but this has not been consistently observed as a motivation amongst cannabis-dependence participants. (Arendt et al., 2007). Alternatively, cannabis use and abuse and major depressive disorder may have common liabilities (e.g. Ottena et al., 2010; Sherva et al., 2016), which may provide an explanation for their observed co-occurrence in the results of this thesis.

7.5 How Do Genetic Influences Contribute to Transitions in Cannabis Use?

The use of a twin sample to study the genetic contribution to transitions in drug use was one of the strengths of this research. Doing so demonstrated that heritability estimates were similar for age at first opportunity to use cannabis and development of lifetime abuse and/or dependence. This is in contrast to existing research suggesting that heritability increases as drug use progresses (Lynskey et al., 2010). Heritability of age of opportunity to use cannabis has rarely been studied previously, and the majority of this previous research focuses on cannabis initiation. This may explain the inconsistency with previous findings.

Additive genetic effects were estimated to account for 66% of the variance in age of opportunity to use cannabis. It has previously been shown that genetic effects influence putatively environmental factors (Kendler and Baker, 2007), of which opportunity to use cannabis is one. Given that genetic effects acting on age of opportunity to use cannabis cannot be drug related, as use has not been initiated at this stage, the overlap in effects must arise from influences that are unrelated to drug metabolism.

A substantial correlation (0.42) was observed between additive genetic influences on age of opportunity to use cannabis and those on cannabis abuse and/or dependence. Many studies have reported a genetic correlation between different stages of drug use. In tobacco research genetic correlation has been identified between i) initiation and amount smoked/cessation (Broms et al., 2006), ii) age of onset and cigarette consumption/smoking persistence (Morley et al., 2007), and iii) cigarette consumption and smoking persistence (Morley et al., 2007). In alcohol research genetic correlations have been identified between: (i) age of initiation and frequency of use/problem use (Pagan et al., 2006); (ii) age of first alcohol use and alcohol dependence (Sartor et al., 2009b); (iii) frequency of use and problem use (Pagan et al., 2006); (iv) heaviness of alcohol use and alcohol abuse/dependence (Grant et al., 2009); and (v) alcohol use disorder and remission (McCutcheon et al., 2012). For cannabis, a genetic correlation has only been identified between number of times of lifetime cannabis use and dependence (Sartor et al., 2010). The analyses in this thesis are the first study to identify a genetic correlation between opportunity to use and later SUD.

The genetic correlation observed between age of opportunity and cannabis abuse and/or dependence was high (r=.42) compared to the majority of existing research. No previous studies have considered age of opportunity, but there has been focus on age of initiation. For alcohol, the genetic correlation between age at initiation and problem use has been estimated as low as .15 and .29 for males and females respectively (Pagan et al., 2006), and as high as .59 for the association between age of first alcohol use and alcohol dependence (Sartor et al., 2009b). The differences in these estimates may reflect differences in the definition of problem use, with only the higher correlation observed using the DSM-IV definition of alcohol dependence. An extremely high genetic correlation of 0.98 has been observed between number of instances of lifetime cannabis use and cannabis dependence (Sartor et al., 2010). The correlation found in this thesis is concordant with the higher estimates previously observed in the literature, but previous findings suggest there is potential for the correlation to be lower should a different measure of problem use be utilised.

The best fitting bivariate twin model was one which equated additive genetic and unique environmental parameters and correlations between age of opportunity and abuse and/or dependence for males and females, which suggests there are not sex differences in the genetic factors acting on these phenotypes, or in the magnitude of the additive genetic effect. This is in contrast to much of the existing literature, which has commonly observed sex differences in genetic correlation between stages of drug use when these were tested for. Correlations in previous studies were observed to be higher in males than for females in all cases (Broms et al., 2006; Morley et al., 2007; Pagan et al., 2006; McCutcheon et al., 2012). However, the present findings are in accordance with the only other study to look at genetic correlations in cannabis use (Sartor et al., 2010).

Existing research has suggested that a proportion of the genetic factors underlying SUDs are not specific to individual drugs (Kendler et al., 2003; Sartor et al., 2010), and the genetic overlap observed within this thesis may represent factors that provide a general liability to drug use and SUDs. Much of the non-specificity of genetic influences on SUDs may result from shared influences on the earlier stages of drug use, with more specific influences (such as those related to metabolism, for example) associated with the later, problematic stages of use.

The results also indicated that genetic effects do not consistently influence

the speed of transition. The inclusion of the novel transition from initiation to subsequent use of cannabis identified a transition that had no observed variance attributable to additive genetic effects. The majority of the variance in this transition was attributable to unique environmental effects. It is important to note that, within twin modelling, estimates of the unique environment also include measurement error (Plomin et al., 2013). The study of the transition to subsequent cannabis use is a novel contribution to the literature, and consequently there is no existing research against which to compare these results. As the subsequent use measure is novel the reliability and validity of it is unknown, and these findings therefore require replication to determine the reliability and validity of the measure. If the measure of subsequent use has high unreliability, additive genetic influences may be masked.

If not due to measurement error, determining what the unique environmental effects could be comprised instance requires speculation. Potential sources of variation include parenting, which can contribute to non-shared environmental sources of variation if twins are treated or perceive themselves to have been treated differently (Plomin et al., 2013). One study of the effect of non-shared environment on delinquent behaviour identified maternal disengagement as a source of such variation (Beaver, 2008), which could plausibly influence this early stage of drug use. The findings highlight the complexity of influences on progression in drug use, and the utility of genetically informative designs to provide important information on the effect of the environment as well as genetics.

7.6 What is the Relationship Between Speed of Early Transitions in Cannabis Use and Later SUDs?

Both earlier opportunity to use cannabis and faster transition from initiation to subsequent use of cannabis were associated with increased likelihood of cannabis daily use, and likelihood of abuse and/or dependence. Age of opportunity alone was associated with likelihood of cannabis related treatment-seeking. An approximately linear relationship was observed between age of cannabis use opportunity and likelihood of progressing to daily cannabis use (OR 2.06 95% CI 1.36-3.11 and 1.67 95% CI 1.18-2.37 for under 14 and 15-16 respectively) or cannabis abuse and/or

dependence (OR 1.91 95% CI 1.33-2.74, 1.86 95% CI 1.40-2.48 and 1.51 95% CI 1.08-2.11 for under 14, 15-16 and 17 and over respectively). Only those aged 14 and under at first opportunity to use cannabis were at increased likelihood of treatment seeking (OR 2.65, 95% CI 1.51-2.64). For speed of transition from initiation to subsequent use, only the fastest transition speed was associated with progressing to daily cannabis use (OR 2.18, 95% CI 1.42-3.35), but a linear relationship with speed was indicated for progression to cannabis abuse and/or dependence (OR 2.73 95% CI 1.92-3.90 and 1.53 95% CI 1.10-2.11 for transition within a week and within three months respectively). There were no associations with treatment seeking. These findings broadly reflect previous research identifying an association between faster transition to initiation of cannabis use and later cannabis dependence (Swift et al., 2008; Grant and Dawson, 1998; Fergusson and Horwood, 1997). Additionally, these results indicate that those who go on to develop SUDs are at heightened risk for problematic outcomes from the earliest stages of cannabis involvement.

The key implication of this finding is that individuals making faster early transitions in drug use may already be exposed to influences that will increase their likelihood of later negative drug use outcomes. This suggests that some of the contribution to SUD risk liability is in evidence at the very earliest stage of drug use, and cannot be attributed to the pharmacological effects of the drug. The association between age of opportunity to use cannabis and later SUDs may reflect the genetic correlation between these two phenotypes. For the progression to subsequent use, for which no additive genetic effect was observed, genetics are unlikely to provide an explanation.

The association to later SUDs likely reflects a combination of individual and contextual factors that surround the rapid escalation. One of these factors may be availability. Although unmeasured in the present study, drug availability has previously been proposed as an explanation for changes in the speed of transitions in drug use (Mills et al., 2004). For cannabis, factors in the environment linked to the availability of the drug have been found to account for part of the association between cannabis initiation and the development of cannabis abuse (Gillespie et al., 2009). Research has not yet explored which factors are associated with higher cannabis availability in adolescence, but speed of transition to subsequent cannabis use may act as a proxy measure of this and a combination of other unmeasured factors that increase liability to SUDs.

7.7 Consistency Across Drug Classes? Testing Speed of Transition in Heroin Use

Including heroin-using populations in the study provided a test of whether speed of transition is a relevant phenotype in drugs other than cannabis. This extended the scientific scope of my studies. As stated previously, heroin use is much less prevalent in the general population than cannabis use (UNODC, 2014), but has associated health and dependence risks that are more severe, especially when administration of the drug is through injecting (Strang et al., 1998). Very little existing research has explored speed of transitions in heroin use but the available literature highlights the importance of considering route of administration (see Chapter 1, Section 1.5).

One reason that it was interesting to test the association between transition speed and later outcomes in heroin use was due to the nature of the drug itself. It has previously been observed that, compared to individuals who inject other drugs (e.g., methamphetamine), those injecting heroin have a faster progression from initiation of use (through injecting) to regular use, and a greater severity of dependence (O'Keefe et al., 2016). Additionally, populations of heroin users often have high levels of childhood abuse and neglect (Darke, 2011). Consequently it was possible that findings relating to transition speed from populations using cannabis may differ in heroin using populations. In the sample in which the relationship between speed of early heroin use transitions and later outcomes was tested, only age of opportunity to use heroin was associated with later outcomes. It may be that heroin pharmacology, or the effect of other unmeasured life course factors, mitigate the relationship between speed of transition and later outcomes.

Overdose was included as an outcome as it is the main cause of mortality amongst opiate users (Degenhardt et al., 2011), and even non-fatal overdose has associated health effects that may have a long-term influence (Warner-Smith et al., 2001). Suicidality and unsafe injecting practices (Bogdanowicz et al., 2016), and lower socioeconomic status and being on the edge of the workforce (Amundsen, 2015), have been found to be associated with overdose mortality. Additionally, non-fatal overdose has been found to be associated with higher levels of use of heroin and other drugs (Darke et al., 2014). However, the association in the present study with earlier opportunity to use indicates that factors contributing to earlier

opportunity may also contribute to overdose risk. Identifying what these factors are, and how they increase the liability to overdose, will help to explain variation in overdose risk and outcome amongst heroin users and may be useful for improving interventions.

The literature review (Chapter 1, Section 1.5) identified that, when exploring heroin transitions, there is value in taking into account drug administration factors. Consequently the analyses in this thesis considered the relationship between of IAR and speed of transitions in heroin use. Injection of heroin was associated with a four-fold increase in progression to daily heroin use, compared to smoking and snorting administration. This is a novel finding, and highlights the importance of considering drug specific factors and transitions when applying a multi-stage approach to drug use.

7.8 Implications of Thesis Findings

7.8.1 Prevention and Intervention

The focus of this project on some of the earliest transitions in drug use has identified that those at increased likelihood of dependence have the potential to be identified at the earliest stage of drug use involvement. Existing research had identified age of initiation as a marker for later problematic use (Behrendt et al., 2009; Lynskey et al., 2012; Swift et al., 2008). Research relating to early onset of drug use often calls for earlier detection and intervention (Chen et al., 2009), and the time between drug use initiation and the development of SUDs has been found to be short, leaving only a small window for targeted intervention (Wittchen et al., 2008). The present findings suggest that those who are at risk can be identified before initiating drug use. This raises the prospect of utilising early drug use behaviours to act as markers for early intervention.

Using the results of the present study may facilitate identification of populations who will benefit from interventions. Selective interventions target those groups who are at increased risk of experiencing an outcome (Poznyak et al., 2011). As discussed previously, there are a large number of risk factors associated with SUDs (Hawkins et al., 1992), each providing potential for intervention. Possibly as a result of the difficulty selecting specific risk factors to target, interventions

for substance use tend to take a universal approach (Teesson et al., 2014) or use personality factors as the selective methods (Barrett et al., 2015; Newton et al., 2016). By applying the multi-stage approach and avoiding conflation of estimates across drug use stages, the present analyses have narrowed the scope of targets for intervention. Adolescents who meet criteria for conduct disorder, whose parents have experienced drug problems, who are engaged in weekly tobacco use, or have had the opportunity to use cannabis at younger ages, may benefit from selective interventions aimed at the prevention of problematic cannabis use.

Indicated interventions target high-risk individuals with early signs of the emerging disorder (Poznyak et al., 2011). A recent Cochrane review of brief interventions for substance-using adolescents identified 5 randomised controlled trials, all aimed at adolescents under age 19 who had used alcohol or other drugs (Carney et al., 2016). The evidence that these interventions were effective at reducing drug or alcohol use was rated as low or very low. Given that the proportion of individuals who initiate cannabis use is much higher than the proportion that will progress to problematic, or even regular, cannabis use, it seems likely that using ever-use of a drug as an indicator for intervention will not maximise effectiveness. The results of this thesis provided a potential new indicator for interventions in the form of speed of transition from initiation to subsequent use of cannabis. If replicated in prospective research, the speed of this transition in cannabis use has the potential to act as a clinically useful indicator of those individuals likely to benefit from intervention; similar to a biomarker (Rifai et al., 2006), but easily and cheaply tested by medical professionals through patient self-report. Although the odds ratios obtained indicate that this measure would have low specificity, making it inappropriate for selecting candidates for intensive intervention, brief interventions such as information provision may be more appropriate.

7.8.2 A Measure of Addictive Liability

There is potential to use measures of transition speed as markers of the addictive liability of a drug. It may be expected that the proportion of a drug using population that experience dependence would be an appropriate marker of addictive liability, but there are issues with this measure; as stated throughout this thesis, the prevalence of dependence is commonly driven by factors separate from the drug

itself (Anthony et al., 1994; Ridenour et al., 2005). Another previously proposed measure of addictive liability is speed of transition from development of abuse symptoms to dependence symptoms (Ridenour et al., 2005), but as discussed in Chapter 1 (Section 1.5) there are issues with the use of this transition.

Speed of transition may provide a better marker of addictive liability. As discussed in Chapter 6, the association between injecting IAR and faster transition to daily heroin use is consistent with what is known about the pharmacology of heroin administration, and likely reflects the higher dependence liability of injection over non-injection routes of heroin administration. This consistency indicates that differences in the speed of transition to daily use may be a useful comparator of drug dependence potential. Given that methodology for assessing transition speed is non-invasive and low cost, this avenue warrants further investigation and may be of use for better understanding emerging novel psychoactive substances.

7.8.3 Selection of Controls in Genetic Research

These findings have additional, important implications for future studies of gene variants and heritability of SUDs, and in the choice of controls in case-control studies. The results of this study indicate that only a moderate proportion of genetic influences on cannabis abuse and/or dependence are unique from those acting on age of opportunity to use cannabis. Consequently genetic studies that do not consider their participants' opportunity to use cannabis risk conflating genetic influences across these stages.

Such arguments have previously been made regarding the importance of considering drug use opportunity when looking at the genetics of opiate use. By comparing participants in treatment for opiate dependence with controls sourced from the ATR (individuals not dependent on alcohol or illicit drugs, with significantly lower illicit drug exposure), and separately with nondependent neighbourhood controls (high exposure to illicit drugs, either via use or from residing in environments with widespread drug availability), differences were found in the SNPs associated with heroin dependence (Nelson et al., 2013). When those in treatment were compared with neighbourhood controls, significant associations with gene variants were identified; these associations were not evident when the ATR controls formed the comparison group. Until now the importance for

genetic studies of considering cannabis use opportunity has not been explored, and no genetic studies of problematic cannabis use have considered cannabis use opportunity amongst the control group.

Consequently a key implication of the findings is the necessity of taking into consideration the stage of drug use reached amongst the controls. Existing research has utilised information on the extent of cannabis use in controls (e.g. excluding those who had used cannabis fewer than 6 times) (Hartman et al., 2009), but such issues are not always being taken into consideration (Benyamina et al., 2009). Depending on the research question, and on the development of research identifying genetic overlap between progression to other stages of cannabis use and problematic cannabis use, screening controls not only for opportunity or initiation of cannabis use, but also for frequency of use may have utility in improving cannabis dependence SNP identification in the future.

7.8.4 Non-Injecting Routes of Heroin Administration

The findings of the thesis identified a relationship between IAR and speed of transition to daily heroin use, with those initiating use through injecting likely to have faster progression to daily use. Additionally, the 'IAR injecting' group had a higher proportion of individuals ever using daily, compared to 'IAR non-injecting' administration groups. Non-injecting administration has previously been shown to be associated with less harm, and reduced likelihood of developing dependence (Strang et al., 1998), overdosing (Gossop et al., 1996) and contracting blood-borne viruses (Strang et al., 1998). The increased latency to daily use amongst participants whose IAR is chasing or snorting supports the harm-reduction case for encouraging non-injecting routes of administration (Bridge, 2010; Wodak and McLeod, 2008).

This has implications for drug treatment provision. The shorter window before daily heroin use for those who initiate through injecting implies that there is a shorter time in which intervention to prevent problem development could occur. The need for drug services to attract those whose treatment needs are less immediate, in order to encourage those already injecting to switch to non-injecting, has previously been suggested in the literature (Strang, 1988). It may be that focus needs to shift again to targeting those who have *recently* begun injecting to provide

encouragement to non-injecting administration (or indeed, heroin abstinence). One trialled method where there are at least preliminary data indicating the approach may be effective is in explicit encouragement and enabling of a switch from injecting to 'chasing' through the distribution of foil packets (required for heroin chasing) at needle exchange centres (Pizzey and Hunt, 2008).

Intervening at the treatment service level will not (and could never be expected to) reach the complete population of individuals who have recently initiated heroin use, and therefore further measures may be required. One method of discouraging injecting drug use amongst these populations is to work with those already engaged in injecting drug use. Intervention that alters the perceptions of current injectors, and focuses on the value of dissuading those not currently injecting heroin from beginning to use this route of administration, has been trialled previously with some success (Hunt et al., 1998). Alternatively, changes can be made at the policy level to reduce the incentives around injecting drug use. One idea that has previously been suggested is that of altering policing to penalise the supply of injectable heroin whilst being more tolerant of the supply of heroin that could only be smoked (Strang and King, 1997). The findings of this thesis suggest that doing so has the potential to limit the harms of heroin use and reduce the prevalence of dependence by not only reducing the number of heroin users who progress to daily use, but also to allow a greater window for treatment services to reach heroin users before a daily habit has developed.

7.8.5 Studies of Gene-Environment Interaction

The present study has explored the extent of genetic influences on speed of transitions in cannabis use, and identified a number of external factors associated with progression through the stages of drug use. However, one of the key findings was that associated factors, and the extent to which additive genetic effects influence transitions in drug use, are not consistent across the stages of drug use. The study of SUD aetiology is increasingly recognising that environmental influences vary by genotype, and that the environment alters the effect a gene has on an individual's physiology (GxE) (Plomin et al., 2013). The present findings indicate the potential for GxE to differ at each stage of drug use, given that factors have been identified that are uniquely associated with specific stages of use, and

the genetic correlation between age of opportunity and abuse/dependence was well below 1.0. One study that has looked at the differences in the levels of individual alcohol use has identified an interaction between the *OPRM1 G* allele and parental rule setting (van der Zwaluw et al., 2014). Future research applying a GxExS(tage) approach, whereby the interaction of specific genes and external factors is considered in light of the stage of drug use, may be key for understanding what underlies individual variation in the progression of drug use.

This approach will have implications for prevention and intervention methods. In drug use, these methods often take into account their target age and specific drug types (Stockings et al., 2016), and recent developments have indicated genotype can underlie differential susceptibility to prevention measures (Brody et al., 2013). Taking the stages of drug use into consideration opens up the potential to develop interventions that specifically target the factors associated with progression through the drug use stages.

7.8.6 Public Health

When studying cannabis use there is a need to consider changes in the drug, both in terms of legal status and chemical composition. Prevalence of cannabis use doubled in the US between 2001 and 2013, although the evidence on whether this has translated into an increase in cannabis use disorders is unclear (Hasin et al., 2015; Grucza et al., 2016). This increase in use may reflect changing attitudes towards cannabis, and in combination with the increasing liberalisation of cannabis control policies (Hopfer, 2014; Shi et al., 2015) may lead to cohort changes in the demographics of cannabis use. Additionally, changes in the administration and chemical composition of cannabis warrant exploration. The strength of cannabis, in terms of the proportion of the content that is composed of tetrahydrocannabinol (THC), which causes many of the "high" effects of cannabis use, has been increasing in recent years (Golick, 2016). Understanding how the THC strength relates to speed of transitions in cannabis use could provide understanding of any increased addictive liability associated with increased strength (see discussion above, Section 7.8.2). A regulated cannabis market (Pacula et al., 2014), in which the THC content of the drug is controlled through legislation and a legal supply market, would be necessary to ensure that forms available were those associated

with slower transitions.

7.9 Methodological Strengths and Limitations

7.9.1 Selection and Testing of Associated Factors

A strength of the study was that a consistent selection of proximal and distal factors was tested for their association with multiple drug use transitions. One of the limitations identified in the literature on age of initiation, and which also applies to the review of research on speed of transitions (see Chapter 1) was that the factors tested for association with speed of transition varied widely, and in some cases did not take into account factors beyond those present early in childhood (Storr et al., 2011). The use of the SSAGA interview (Bucholz et al., 1994) provided information on a wide range of potential covariates from childhood, adolescence and into adulthood. As discussed above, this approach revealed a number of novel findings on the consistency of, and changes in, influences on transition speed across transitions.

7.9.2 The Use of Multiple Samples

The study utilised multiple samples to address different questions. Cannabis use has a relatively high base-rate in the general population, and could therefore be studied using a non-clinical sample. The use of a relatively large (N = 3824) twin sample provided sufficient power to conduct genetically informative analyses and test a range of hypotheses in epidemiological analyses. Although necessarily smaller, samples incorporating individuals already using heroin allowed the study of outcomes in a drug that has a low base rate in general population samples.

7.9.3 Self-Report

All data were collected through self-report. Self-report has been recommended as the gold standard method for collecting data on drug use (Wagner and Anthony, 2002), and all studies guaranteed participant confidentiality. However, despite this there is potential that participants would have provided inaccurate information on

topics they felt uncomfortable discussing. Given that use of cannabis is illegal in the countries data were collected in (at time of data collection and thesis writing), some participants in the ATR study may have incorrectly reported on their drug use. However, given the high prevalence of self-reported lifetime cannabis use (68.5%), it seems unlikely that this was a problem.

For the heroin samples, which were comprised of individuals already selected for their heroin use, participants can be expected to be less likely to withhold information on use of the drug. A review of the reliability and validity of self-report in populations of injecting drug users found no differences in report accuracy between those in treatment and those not in treatment, good reliability and validity for self-report of amount and frequency of drug use in a number of studies, and high concordance between self-reported drug use and independent measures (Darke, 1998). Only one study suggests an issue which may affect the results of the present study, which is the estimated amount of heroin used was higher when the interview was conducted by a researcher, compared to when conducted by peer interviewer (Davies and Baker, 1987). Therefore, despite the limitations of the method (and aside from the unfeasible idea of regular drug testing of an adolescent or young adult cohort), self-report is the best method available for measuring onset of the stages of drug use.

7.9.4 Recall Bias

All data were collected retrospectively, introducing the potential for recall bias. This refers to the phenomenon whereby the exposure is recalled differentially between groups who did and did not experience the outcome of interest, either as a result of the outcome affecting recall of the exposure or leading to exaggeration amongst those who experienced it (Hotopf, 2003; Hennekens and Buring, 1987). For example, it is feasible that those who have developed SUDs may have spent more time reflecting back upon their early drug use than those who did not experience problems, leading to differential recall of the speed of their early transitions in drug use.

However, research into the reliability of recall of first use of cannabis, alcohol and tobacco, tested by asking participants within a longitudinal study the same questions at a later time point, indicated the there is good recall of these

milestones (intraclass correlation for recall 4+ years after first asking 0.67; intraclass correlation 0.73 for recall of cannabis items) (Johnson and Mott, 2001). Moderate reliability has been found for self-reported age of drug, alcohol and tobacco onset in an 11-year prospective cohort study, with the age at which onset was associated found to have no effect on the relationship to later drug outcomes (Parra et al., 2003). A moderate correlation of r=0.73 (95% CI: 0,71-0.74) and r=0.76 (95% CI: 0.74-0,78) for males and females respectively was observed in a study of those asked about age of tobacco initiation at age 18-19, and again at 20-22 (Huerta et al., 2005). A study of participants asked about cannabis initiation 10 years after being asked at baseline found 99.5% of participants reported an age of onset that matched their baseline reports (Shillington et al., 1995). Age of alcohol use had a moderate correlation with report 2 years after baseline assessment (0.65) (Greenfield et al., 2014). In a sample longitudinally followed from 6 to age 32, cannabis use in adolescence was consistently reported by 64% of the sample, with only 9% inconsistently reporting their age of onset (Ensminger et al., 2007).

Much prior research utilised retrospective report of age of initiation and transitions in cannabis and heroin use (Barrio et al., 2001; Grant et al., 2010; Kendler et al., 2003, 2008; Kendler and Prescott, 1998; Lopez-Quintero et al., 2011; Ridenour et al., 2005; Sartor et al., 2013a; van Etten and Anthony, 1999; Wagner and Anthony, 2007; Winters and Lee, 2008; Woodcock et al., 2015). Consequently retrospective recall can be a said to an appropriate method of assessing early drug use behaviours, but results would benefit from replication in prospective studies.

7.9.5 The Use of Twin Samples

The majority of analyses in this thesis are based on a general population twin sample. It may be expected that twins will differ from non-twins, which raises questions as to the extent to which findings from twin samples can be applied to the general, non-twin population. However, researchers have consistently shown that twins sampled from the general population are representative of that population; particularly in relation to the prevalence of mental health problems, which have been shown to not differ between twin and non-twin populations (Kendler et al., 1996). Additionally twin environmental similarity has been shown to not be associated with mental health outcomes (Kendler et al., 1993), which means

we should not expect twin populations to differ from non-twin samples on such measures. Publications in multiple journals and fields have utilised a twin sample as equivalent to a general population sample (Åkerstedt et al., 2015; Ropponen and Svedberg, 2014; Valdés et al., 2014; Waldron et al., 2014; Wulf-Johansson et al., 2013), with many epidemiological analyses published from Scandinavian twin cohorts. Consequently, whilst the use of a twin sample has potential to be considered a limitation, it is appropriate for the analyses in this thesis.

The Equal Environments Assumption

As stated in Chapter 2 (Section 2.5.5), one of the key assumptions of twin modelling is that, despite the differences in genetic similarity, environmental influences will be equally correlated in both monozygotic and dizygotic twins. This is important as estimates of heritability will be incorrectly inflated if there is greater similarity in MZ environments, as the heritability estimate is based on the principle that a greater correlation with an outcome in MZ twins compared to DZ twins' results will be due to the increased genetic similarity of MZ twins (Kendler et al., 1993). Analyses conducted on twins who reported equal and unequal environments demonstrated that equality of environment did not affect estimates for cannabis abuse and/or dependence. These findings are consistent with existing research demonstrating that this is not a limitation in analyses of twin data (Kendler and Gardner, 1998; Kendler et al., 1993; LoParo and Waldman, 2014; Plomin et al., 1976).

7.10 Future Research Directions

The study has highlighted the value of applying a multi-stage approach to drug use, and of considering variation in speed of transition. Doing so has led to interesting conclusions about the relationship between speed of early transitions and later drug use outcomes, and the ways in which influences on drug use alter across stages and transitions. These findings have also raised a number of questions that would benefit from exploration in future research.

The analyses in this thesis were exploratory, applying the relatively novel approach of considering speed of transitions in drug use in cross-sectional samples to gain a broad understanding of the relevance of transitions in drug use. Having

identified associations between speed of early transitions in drug use and later drug use outcomes, and having identified that there are both consistent and stage specific influences on progression in drug use, there would be value in exploring this topic in more depth. Using prospective cohorts to test whether there is replication of the associations between early opportunity to use cannabis or heroin and later drug use outcomes, and between faster transition to subsequent cannabis use and later cannabis outcomes, would be necessary before progressing to use of these transitions as markers for intervention. There is also a need to consider the effect of individual factors on the speed of transitions, and potentially to test mediation of the relationship to later drug use outcomes. Important targets to incorporate in future studies of transitions are suggested in research from the alcohol field. Social context of drug use (Degenhardt et al., 2015), motives for use (Littlefield et al., 2010), and personality changes across adolescence (Hicks et al., 2012) have been found to affect problematic drinking in adolescence. Finally, given that the early stages of drug use and progression are so linked to adolescence, there is value in considering drug use in the context of adolescent transitions. These include education completion and moving out of parents home (Eisenberg et al., 2015), and exploring these in terms of the effect of these on drug use and the associations between earlier drug use and reaching these stages.

The findings in this thesis have highlighted that both genetic and environmental factors contribute to progression in drug use, and as suggested above, there is scope to develop a GxExS(tage) approach to the study of drug use. With regard to the environmental exposures selected for study, there are an extensive number of potential exposures that can be combined with the plethora of gene candidates. Sher et al. (2010) have highlighted that the unavailability of information on the number of possible environmental influences means that there is no "environome" to compare with the genome. Distal factors that are thought to have a long-term effect (such as childhood family environment) can be differentiated from proximal exposures (such as availability), and studies of G×ExS in adolescent drug use may benefit from systematically classifying environment in this way. Additionally, for SUDs as with other psychiatric disorders, despite years of candidate gene studies there are still few replicated associations (Duncan and Keller, 2011; Bosker et al., 2011; Reich et al., 1999; Tabor et al., 2002). Focussing research on combinations of genes or gene systems through the use of polygenic risk scores have produced

promising results (Belsky et al., 2013; McGeary et al., 2012; Salvatore et al., 2014), and may be a more efficient strategy for understanding G×ExS in drug use. The application of these to stage transitions has the potential to elucidate the complexities underlying the development of drug dependence.

The findings of this thesis indicate that those who progress more rapidly to cannabis dependence are likely to have additional co-occurring drug use and dependency. Those who use cannabis have been found to be more likely to have opportunities to use drugs such as heroin (Strang and McCambridge, 2005). It has also been observed that amongst those who use multiple drugs, the age of onset of these drugs has declined across birth cohorts, with the result that those who initiate cannabis use in their teens are likely to be exposed to a broad range of drug use (Darke et al., 2012). As discussed previously in this thesis, there is common genetic liability to drug dependencies and there are proposed mechanisms underlying the co-occurrence of cannabis and tobacco use. However, beyond this there is little understanding of the ways in which multiple dependencies develop, the levels of problem use of other drugs amongst those diagnosed as dependent on a specific drug, and the implications that this has for prevention and treatment of drug dependencies. Pursuing the following avenues of research could make valuable contributions to the literature

1. identifying the extent to which adolescents experience co-occurring problematic drug use (frequent or heavy use of alcohol, cannabis or other drugs)
2. studying whether this use is stable or whether levels of use alter with age
3. exploring the proximal and distal factors associated with multiple problematic drug use
4. identifying how this impacts on later life social, mental health and drug use outcomes

Bibliography

Agosti, V., Nunes, E., and Levin, F. (2002). Rates of Psychiatric Comorbidity Among U.S. Residents with Lifetime Cannabis Dependence. *The American Journal of Drug and Alcohol Abuse*, 28(4):643–652.

Agrawal, A., Budney, A. J., and Lynskey, M. T. (2012). The co-occurring use and misuse of cannabis and tobacco: a review. *Addiction*, 107(7):1221–1233.

Agrawal, A., Grant, J. D., Waldron, M., Duncan, A. E., Scherrer, J. F., Lynskey, M. T., Madden, P. A. F., Bucholz, K. K., and Heath, A. C. (2006). Risk for initiation of substance use as a function of age of onset of cigarette, alcohol and cannabis use: Findings in a Midwestern female twin cohort. *Preventive Medicine*, 43(2):125–128.

Agrawal, A. and Lynskey, M. T. (2006). The genetic epidemiology of cannabis use, abuse and dependence. *Addiction (Abingdon, England)*, 101(6):801–812.

Agrawal, A. and Lynskey, M. T. (2008). Are there genetic influences on addiction: evidence from family, adoption and twin studies. *Addiction*, 103(7):1069–81.

Agrawal, A., Madden, P. A. F., Martin, N. G., and Lynskey, M. T. (2013). Do early experiences with cannabis vary in cigarette smokers? *Drug and Alcohol Dependence*, 128(3):255–259.

Agrawal, A., Neale, M. C., Prescott, C. A., and Kendler, K. S. (2004). Cannabis and

other illicit drugs: comorbid use and abuse/dependence in males and females. *Behav Genet*, 34(3):217–28.

Agrawal, A., Silberg, J. L., Lynskey, M. T., Maes, H. H., and Eaves, L. J. (2010). Mechanisms underlying the lifetime co-occurrence of tobacco and cannabis use in adolescent and young adult twins. *Drug and Alcohol Dependence*, 108(1–2):49–55.

Åkerstedt, T., Knutsson, A., Narusyte, J., Svedberg, P., Kecklund, G., and Alexanderson, K. (2015). Night work and breast cancer in women: a Swedish cohort study. *BMJ Open*, 5(4):e008127.

Altman, D. G. (1994). Problems in dichotomizing continuous variables. *American Journal of Epidemiology*, 139(4):442–445.

American Psychiatric Association (2000). *Diagnostic and statistical manual of mental disorders: DSM-IV-TR*. American Psychiatric Association, Washington, DC, 4th ed., text revision edition.

American Psychiatric Association, American Psychiatric Association, and DSM-5 Task Force (2013). *Diagnostic and statistical manual of mental disorders: DSM-5.* American Psychiatric Association.

Amundsen, E. J. (2015). Drug-related causes of death: Socioeconomic and demographic characteristics of the deceased. *Scandinavian Journal of Public Health*, 43(6):571–579. WOS:000360391600003.

Anthony, J. (2006). The Epidemiology of Cannabis Dependence. In Roffman, R. A. and Stephens, R. S., editors, *Cannabis Dependence: Its Nature, Consequences, and Treatment*, pages 58–105. Cambridge University Press, New York.

Anthony, J. C., Warner, L. A., and Kessler, R. C. (1994). Comparative epidemiology of dependence on tobacco, alcohol, controlled substances, and inhalants: Basic findings from the national comorbidity survey. *Experimental and Clinical Psychopharmacology*, 2(3):244–268.

Arendt, M., Rosenberg, R., Fjordback, L., Brandholdt, J., Foldager, L., Sher, L., and Munk-Jørgensen, P. (2007). Testing the self-medication hypothesis of

depression and aggression in cannabis-dependent subjects. *Psychological Medicine*, 37(07):935–945.

Barker, E. D. and Maughan, B. (2009). Differentiating Early-Onset Persistent Versus Childhood-Limited Conduct Problem Youth. *American Journal of Psychiatry*, 166(8):900–908.

Barrett, E. L., Newton, N. C., Teesson, M., Slade, T., and Conrod, P. J. (2015). Adapting the personality-targeted Preventure program to prevent substance use and associated harms among high-risk Australian adolescents. *Early Intervention in Psychiatry*, 9(4):308–315.

Barrio, G., De La Fuente, L., Lew, C., Royuela, L., Bravo, M. J., and Torrens, M. (2001). Differences in severity of heroin dependence by route of administration: the importance of length of heroin use. *Drug and Alcohol Dependence*, 63(2):169–177.

Beaver, K. M. (2008). Nonshared Environmental Influences on Adolescent Delinquent Involvement and Adult Criminal Behavior*. *Criminology*, 46(2):341–369.

Becker, B., Wagner, D., Gouzoulis-Mayfrank, E., Spuentrup, E., and Daumann, J. (2010). The impact of early-onset cannabis use on functional brain correlates of working memory. *Progress in Neuro-Psychopharmacology and Biological Psychiatry*, 34(6):837–845.

Behrendt, S., Beesdo-Baum, K., Hofler, M., Perkonigg, A., Buhringer, G., Lieb, R., and Wittchen, H. U. (2012). The relevance of age at first alcohol and nicotine use for initiation of cannabis use and progression to cannabis use disorders. *Drug and alcohol dependence*, 123(1-3):48–56.

Behrendt, S., Wittchen, H. U., Hofler, M., Lieb, R., and Beesdo, K. (2009). Transitions from first substance use to substance use disorders in adolescence: is early onset associated with a rapid escalation? *Drug and alcohol dependence*, 99(1-3):68–78.

Bellera, C. A., MacGrogan, G., Debled, M., Lara, C. T. d., Brouste, V., and Mathoulin-Pélissier, S. (2010). Variables with time-varying effects and the Cox model: Some

statistical concepts illustrated with a prognostic factor study in breast cancer. *BMC Medical Research Methodology*, 10(1):20.

Belsky, D. W., Moffitt, T. E., Baker, T., Evans, J., Harrington, H., Houts, R., Meier, M., Sugden, K., Williams, B., Poulton, R., and Caspi, A. (2013). Polygenic risk and the developmental progression to heavy, persistent smoking and nicotine dependence: Evidence from a 4-decade longitudinal study. *JAMA Psychiatry*, 70(5):534–542.

Benjet, C., Borges, G., Medina-Mora, M. E., and Méndez, E. (2013). Chronic childhood adversity and stages of substance use involvement in adolescents. *Drug and Alcohol Dependence*, 131(1–2):85–91.

Benyamina, A., Bonhomme-Faivre, L., Picard, V., Sabbagh, A., Richard, D., Blecha, L., Rahioui, H., Karila, L., Lukasiewicz, M., Picard, V., Marill, C., and Reynaud, M. (2009). Association between ABCB1 C3435t polymorphism and increased risk of cannabis dependence. *Progress in Neuro-Psychopharmacology and Biological Psychiatry*, 33(7):1270–1274.

Best, D., Gross, S., Manning, V., Gossop, M., Witton, J., and Strang, J. (2005). Cannabis use in adolescents: the impact of risk and protective factors and social functioning. *Drug and Alcohol Review*, 24(6):483–488.

Bogdanowicz, K. M., Stewart, R., Chang, C.-K., Downs, J., Khondoker, M., Shetty, H., Strang, J., and Hayes, R. D. (2016). Identifying mortality risks in patients with opioid use disorder using brief screening assessment: Secondary mental health clinical records analysis. *Drug and Alcohol Dependence*, 164:82–88. WOS:000378468800011.

Boker, S. M., Neale, M. C., Maes, H. H., Wilde, M. J., Spiegel, M., Brick, T. R., Spies, J., Estabrook, R., Kenny, S., Bates, T. C., Mehta, P., and Fox, J. (2011). OpenMx: An Open Source Extended Structural Equation Modeling Framework. *Psychometrika*, 76(2):306–317.

Bosker, F., Hartman, C., Nolte, I., Prins, B., Terpstra, P., Posthuma, D., van Veen, T., Willemsen, G., DeRijk, R., de Geus, E., Hoogendijk, W., Sullivan, P., Penninx, B., Boomsma, D., Snieder, H., and Nolen, W. (2011). Poor replication of

candidate genes for major depressive disorder using genome-wide association data. *Molecular Psychiatry*, 16:516–32.

Bridge, J. (2010). Route transition interventions: Potential public health gains from reducing or preventing injecting. *International Journal of Drug Policy*, 21(2):125–128.

Brody, G. H., Chen, Y.-f., and Beach, S. R. H. (2013). Differential susceptibility to prevention: GABAergic, dopaminergic, and multilocus effects. *Journal of Child Psychology and Psychiatry, and Allied Disciplines*, 54(8):863–871.

Broms, U., Silventoinen, K., Madden, P. A. F., Heath, A. C., and Kaprio, J. (2006). Genetic Architecture of Smoking Behavior: A Study of Finnish Adult Twins. *Twin Research and Human Genetics*, 9(1):64–72.

Bucholz, K. K., Cadoret, R., Cloninger, C. R., Dinwiddie, S. H., Hesselbrock, V. M., Nurnberger, J., Reich, T., Schmidt, I., and Schuckit, M. A. (1994). A New, Semi-Structured Psychiatric Interview for Use in Genetic Linkage Studies: A Report on the Reliability of the SSAGA. *Journal of Studies on Alcohol and Drugs*, 55(2):149.

Caris, L., Wagner, F. A., Ríos-Bedoya, C. F., and Anthony, J. C. (2009). Opportunities to use drugs and stages of drug involvement outside the United States: Evidence from the Republic of Chile. *Drug and Alcohol Dependence*, 102(1–3):30–34.

Carney, T., Myers, B. J., Louw, J., and Okwundu, C. I. (2016). Brief school-based interventions and behavioural outcomes for substance-using adolescents. In The Cochrane Collaboration, editor, *Cochrane Database of Systematic Reviews*. John Wiley & Sons, Ltd, Chichester, UK.

Chamla, D., Chamla, J. H., Dabin, W., Delin, H., and Rennes, N. (2006). Transition to injection and sharing of needles/syringes: Potential for HIV transmission among heroin users in Chengdu, China. *Addictive Behaviors*, 31(4):697–701.

Chen, C.-Y., Dormitzer, C. M., Bejarano, J., and Anthony, J. C. (2004). Religiosity and the Earliest Stages of Adolescent Drug Involvement in Seven Countries of Latin America. *American Journal of Epidemiology*, 159(12):1180–1188.

Chen, C.-Y., O'Brien, M. S., and Anthony, J. C. (2005a). Who becomes cannabis dependent soon after onset of use? Epidemiological evidence from the United States: 2000–2001. *Drug and Alcohol Dependence*, 79(1):11–22.

Chen, C. Y., Storr, C. L., and Anthony, J. C. (2005b). Influences of parenting practices on the risk of having a chance to try cannabis. *Pediatrics*, 115(6):1631–9.

Chen, C.-Y., Storr, C. L., and Anthony, J. C. (2009). Early-onset drug use and risk for drug dependence problems. *Addictive Behaviors*, 34(3):319–322.

Coffey, C., Lynskey, M., Wolfe, R., and Patton, G. C. (2000). Initiation and progression of cannabis use in a population-based Australian adolescent longitudinal study. *Addiction*, 95(11):1679–1690.

Conway, K. P., Levy, J., Vanyukov, M., Chandler, R., Rutter, J., Swan, G. E., and Neale, M. (2010). Measuring addiction propensity and severity: The need for a new instrument. *Drug and Alcohol Dependence*, 111(1–2):4–12.

Creemers, H. E., Korhonen, T., Kaprio, J., Vollebergh, W. A. M., Ormel, J., Verhulst, F. C., and Huizink, A. C. (2009a). The role of temperament in the relationship between early onset of tobacco and cannabis use: The TRAILS study. *Drug and Alcohol Dependence*, 104(1–2):113–118.

Creemers, H. E., Lier, P. A. C. v., Vollebergh, W. A. M., Ormel, J., Verhulst, F. C., and Huizink, A. C. (2009b). Predicting Onset of Cannabis Use in Early Adolescence: The Interrelation Between High-Intensity Pleasure and Disruptive Behavior. The TRAILS Study [Corrected Version]. *Journal of Studies on Alcohol and Drugs*, 70(6):850.

Danielsson, A.-K., Lundin, A., Agardh, E., Allebeck, P., and Forsell, Y. (2016). Cannabis use, depression and anxiety: A 3-year prospective population-based study. *Journal of Affective Disorders*, 193:103–108. WOS:000369586000015.

Darke, S. (1998). Self-report among injecting drug users: A review. *Drug and Alcohol Dependence*, 51(3):253–263.

Darke, S. (2011). *The Life of the Heroin User*. Cambridge University Press.

Darke, S., Kaye, S., and Torok, M. (2012). Age-related patterns of drug use initiation among polydrug using regular psychostimulant users. *Drug and Alcohol Review*, 31(6):784–789.

Darke, S., Marel, C., Mills, K. L., Ross, J., Slade, T., Burns, L., and Teesson, M. (2014). Patterns and correlates of non-fatal heroin overdose at 11-year follow-up: Findings from the Australian Treatment Outcome Study. *Drug and Alcohol Dependence*, 144:148–152. WOS:000345201500017.

Darke, S., Ross, J., and Kaye, S. (2001). Physical injecting sites among injecting drug users in Sydney, Australia. *Drug and Alcohol Dependence*, 62(1):77–82.

Davies, J. B. and Baker, R. (1987). The Impact of Self-presentation and Interviewer Bias Effects on Self-reported Heroin Use. *British Journal of Addiction*, 82(8):907–912.

Degenhardt, L., Bucello, C., Mathers, B., Briegleb, C., Ali, H., Hickman, M., and McLaren, J. (2011). Mortality among regular or dependent users of heroin and other opioids: a systematic review and meta-analysis of cohort studies. *Addiction (Abingdon, England)*, 106(1):32–51.

Degenhardt, L., Chiu, W.-T., Sampson, N., Kessler, R. C., Anthony, J. C., Angermeyer, M., Bruffaerts, R., de Girolamo, G., Gureje, O., Huang, Y., Karam, A., Kostyuchenko, S., Lepine, J. P., Mora, M. E. M., Neumark, Y., Ormel, J. H., Pinto-Meza, A., Posada-Villa, J., Stein, D. J., Takeshima, T., and Wells, J. E. (2008). Toward a Global View of Alcohol, Tobacco, Cannabis, and Cocaine Use: Findings from the WHO World Mental Health Surveys. *PLoS Med.*, 5(7):e141.

Degenhardt, L., Romaniuk, H., Coffey, C., Hall, W. D., Swift, W., Carlin, J. B., O'Loughlin, C., and Patton, G. C. (2015). Does the social context of early alcohol use affect risky drinking in adolescents? Prospective cohort study. *BMC public health*, 15(1):1137.

Degenhardt, L., Whiteford, H., and Hall, W. D. (2014). The Global Burden of Disease projects: What have we learned about illicit drug use and dependence and their contribution to the global burden of disease? *Drug and Alcohol Review*, 33(1):4–12.

Degenhardt, L., Whiteford, H. A., Ferrari, A. J., Baxter, A. J., Charlson, F. J., Hall, W. D., Freedman, G., Burstein, R., Johns, N., Engell, R. E., Flaxman, A., Murray, C. J., and Vos, T. (2013). Global burden of disease attributable to illicit drug use and dependence: findings from the Global Burden of Disease Study 2010. *The Lancet*, 382(9904):1564–1574.

DeWit, D. J., Hance, J., Offord, D. R., and Ogborne, A. (2000). The Influence of Early and Frequent Use of Marijuana on the Risk of Desistance and of Progression to Marijuana-Related Harm. *Preventive Medicine*, 31(5):455–464.

Dick, D. M., Cho, S. B., Latendresse, S. J., Aliev, F., Nurnberger, J. I., Edenberg, H. J., Schuckit, M., Hesselbrock, V. M., Porjesz, B., Bucholz, K., Wang, J.-C., Goate, A., Kramer, J. R., and Kuperman, S. (2014). Genetic influences on alcohol use across stages of development: GABRA2 and longitudinal trajectories of drunkenness from adolescence to young adulthood. *Addiction Biology*, 19(6):1055–1064.

Ding, Y., He, N., Shoptaw, S., Gao, M., and Detels, R. (2013). Severity of club drug dependence and perceived need for treatment among a sample of adult club drug users in Shanghai, China. *Social Psychiatry and Psychiatric Epidemiology*, 49(3):395–404.

Distel, M. A., Vink, J. M., Bartels, M., van Beijsterveldt, C. E., Neale, M. C., and Boomsma, D. I. (2011). Age moderates non-genetic influences on the initiation of cannabis use: a twin-sibling study in Dutch adolescents and young adults. *Addiction*, 106(9):1658–66.

Do, E. K., Prom-Wormley, E. C., Eaves, L. J., Silberg, J. L., Miles, D. R., and Maes, H. H. (2015). Genetic and Environmental Influences on Smoking Behavior across Adolescence and Young Adulthood in the Virginia Twin Study of Adolescent Behavioral Development and the Transitions to Substance Abuse Follow-Up. *Twin Research and Human Genetics*, 18(01):43–51.

Donovan, J. E. and Molina, B. S. G. (2011). Childhood Risk Factors for Early-Onset Drinking. *Journal of Studies on Alcohol and Drugs*, 72(5):741–751.

Dube, S. R., Miller, J. W., Brown, D. W., Giles, W. H., Felitti, V. J., Dong, M., and Anda, R. F. (2006). Adverse childhood experiences and the association with ever using alcohol and initiating alcohol use during adolescence. *The Journal of Adolescent*

Health: Official Publication of the Society for Adolescent Medicine, 38(4):444.e1–10.

Duncan, L. E. and Keller, M. C. (2011). A critical review of the first 10 years of candidate gene-by-environment interaction research in psychiatry. *The American Journal of Psychiatry*, 168(10):1041–1049.

Efron, B. (1977). The Efficiency of Cox's Likelihood Function for Censored Data. *Journal of the American Statistical Association*, 72(359):557–565.

Eisenberg, M. E., Spry, E., and Patton, G. C. (2015). From Emerging to Established Longitudinal Patterns in the Timing of Transition Events Among Australian Emerging Adults. *Emerging Adulthood*, 3(4):277–281.

Elkins, I., McGue, M., and Iacono, W. G. (2007). Prospective effects of attention-deficit/hyperactivity disorder, conduct disorder, and sex on adolescent substance use and abuse. *Archives of General Psychiatry*, 64(10):1145–1152.

Ensminger, M. E., Juon, H.-S., and Green, K. M. (2007). Consistency between adolescent reports and adult retrospective reports of adolescent marijuana use: Explanations of inconsistent reporting among an African American population. *Drug & Alcohol Dependence*, 89(1):13–23.

Faul, F., Erdfelder, E., Buchner, A., and Lang, A.-G. (2007). G*power 3: A flexible statistical power analysis program for the social, behavioral, and biomedical sciences. *Behavior Research Methods*, 39:175–191.

Feingold, D., Weiser, M., Rehm, J., and Lev-Ran, S. (2014). The association between cannabis use and mood disorders: A longitudinal study. *Journal of Affective Disorders*, 172C:211–218.

Fergusson, D. M. and Horwood, L. J. (1997). Early onset cannabis use and psychosocial adjustment in young adults. *Addiction*, 92(3):279–296.

Fergusson, D. M., Lynskey, M. T., and Horwood, L. J. (1993). Conduct Problems and Attention Deficit Behaviour in Middle Childhood and Cannabis Use by Age 15. *Australian and New Zealand Journal of Psychiatry*, 27(4):673–682.

Fisher, L. D. and Lin, D. Y. (1999). Time-Dependent Covariates in the Cox Proportional-Hazards Regression Model. *Annual Review of Public Health*, 20(1):145–157.

Fowler, T., Lifford, K., Shelton, K., Rice, F., Thapar, A., Neale, M. C., McBride, A., and van den Bree, M. B. M. (2007). Exploring the relationship between genetic and environmental influences on initiation and progression of substance use. *Addiction (Abingdon, England)*, 102(3):413–422.

Fuller, E., Jotangia, D., and Farrell, M. (2009). Drug misuse and Dependence. Technical report, The NHS Information Centre for health and social care.

Gage, S. H., Hickman, M., Heron, J., Munafò, M. R., Lewis, G., Macleod, J., and Zammit, S. (2015). Associations of cannabis and cigarette use with depression and anxiety at age 18: findings from the Avon Longitudinal Study of Parents and Children. *PloS One*, 10(4):e0122896.

Galéra, C., Bouvard, M. P., Melchior, M., Chastang, J. F., Lagarde, E., Michel, G., Encrenaz, G., Messiah, A., and Fombonne, E. (2010). Disruptive symptoms in childhood and adolescence and early initiation of tobacco and cannabis use: The Gazel Youth study. *European Psychiatry*, 25(7):402–408.

Gillespie, N. A., Kendler, K. S., Prescott, C. A., Aggen, S. H., Gardner, C. O., Jacobson, K., and Neale, M. C. (2007). Longitudinal modeling of genetic and environmental influences on self-reported availability of psychoactive substances: alcohol, cigarettes, marijuana, cocaine and stimulants. *Psychological Medicine*, 37(07):947–959.

Gillespie, N. A. and Martin, N. G. (2005). *Multivariate Genetic Analysis*. John Wiley & Sons, Ltd.

Gillespie, N. A., Neale, M. C., and Kendler, K. S. (2009). Pathways to cannabis abuse: a multi-stage model from cannabis availability, cannabis initiation and progression to abuse. *Addiction*, 104(3):430–438.

Golick, J. (2016). Shifting the Paradigm: Adolescent Cannabis Abuse and the Need for Early Intervention. *Journal of Psychoactive Drugs*, 48(1):24–27.

González-Sáiz, F., Domingo-Salvany, A., Barrio, G., Sánchez-Niubó, A., Brugal, M. T., de la Fuente, L., and Alonso, J. (2009). Severity of Dependence Scale as a Diagnostic Tool for Heroin and Cocaine Dependence. *European Addiction Research*, 15(2):87–93.

González-Saiz, F., Lozano, Ó. M., Ballesta, R., Silva, T., Brugal, M. T., Bilbao, I., Barrio, G., Domingo-Salvany, A., Bravo, M. J., De la Fuente, L., and del Proyecto ItÍnere, G. (2008). Validity of the Severity of Dependence Scale (SDS) Construct Applying the Item Response Theory to a Non-Clinical Sample of Heroin Users. *Substance Use & Misuse*, 43(7):919–935.

Gossop, M., Darke, S., Griffiths, P., Hando, J., Powis, B., Hall, W., and Strang, J. (1995). The Severity of Dependence Scale (SDS): psychometric properties of the SDS in English and Australian samples of heroin, cocaine and amphetamine users. *Addiction*, 90(5):607–614.

Gossop, M., Griffiths, P., Powis, B., Williamson, S., and Strang, J. (1996). Frequency of non-fatal heroin overdose: survey of heroin users recruited in non-clinical settings. *BMJ*, 313:402.

Grant, B. and Dawson, D. A. (1998). Age of onset of drug use and its association with dsm-iv drug abuse and dependence: Results from the national longitudinal alcohol epidemiologic survey. *Journal of Substance Abuse*, 10:163–173.

Grant, J. D., Agrawal, A., Bucholz, K. K., Madden, P. A. F., Pergadia, M. L., Nelson, E. C., Lynskey, M. T., Todd, R. D., Todorov, A. A., Hansell, N. K., Whitfield, J. B., Martin, N. G., and Heath, A. C. (2009). Alcohol consumption indices of genetic risk for alcohol dependence. *Biological Psychiatry*, 66(8):795–800.

Grant, J. D., Lynskey, M. T., Madden, P. a. F., Nelson, E. C., Few, L. R., Bucholz, K. K., Statham, D. J., Martin, N. G., Heath, A. C., and Agrawal, A. (2015). The role of conduct disorder in the relationship between alcohol, nicotine and cannabis use disorders. *Psychological Medicine*, 45(16):3505–3515.

Grant, J. D., Lynskey, M. T., Scherrer, J. F., Agrawal, A., Heath, A. C., and Bucholz, K. K. (2010). A cotwin-control analysis of drug use and abuse/dependence risk associated with early-onset cannabis use. *Addictive Behaviors*, 35(1):35–41.

Grant, J. D., Scherrer, J. F., Lynskey, M. T., Lyons, M. J., Eisen, S. A., Tsuang, M. T., True, W. R., and Bucholz, K. K. (2006). Adolescent alcohol use is a risk factor for adult alcohol and drug dependence: evidence from a twin design. *Psychol Med*, 36(1):109–18.

Greenfield, T. K., Nayak, M. B., Bond, J., Kerr, W. C., and Ye, Y. (2014). Test–Retest Reliability and Validity of Life-Course Alcohol Consumption Measures: The 2005 National Alcohol Survey Follow-Up. *Alcoholism: Clinical and Experimental Research*, 38(9):2479–2487.

Griffiths, P., Gossop, M., Powis, B., and Strang, J. (1993). Reaching hidden populations of drug users by privileged access interviewers: methodological and practical issues. *Addiction (Abingdon, England)*, 88(12):1617–1626.

Griffiths, P., Gossop, M., Powis, B., and Strang, J. (1994). Transitions in patterns of heroin administration: a study of heroin chasers and heroin injectors. *Addiction (Abingdon, England)*, 89(3):301–309.

Grucza, R., Agrawal, A., Krauss, M., Bongu, J., Plunk, A., Cavazos-Rehg, P., and Bierut, L. (2016). Declining prevalence of marijuana use disorders among adolescents in the united states, 2002 to 2013. *Journal of Academic Child and Adolescent Psychiatry*, 55:487–494.

Haas, A. L. and Peters, R. H. (2000). Development of substance abuse problems among drug-involved offenders: evidence for the telescoping effect. *Journal of Substance Abuse*, 12(3):241–253.

Hansen, S. E. (2007). CATI Sample Management Systems. In Lepkowski, J. M., Tucker, C., Brick, J. M., Leeuw, E. D. d., Japec, L., Lavrakas, P. J., Link, M. W., and Sangster, R. L., editors, *Advances in Telephone Survey Methodology*, pages 340–358. John Wiley & Sons, Inc.

Hartman, C. A., Hopfer, C. J., Haberstick, B., Rhee, S. H., Crowley, T. J., Corley, R. P., Hewitt, J. K., and Ehringer, M. A. (2009). The association between cannabinoid receptor 1 gene (CNR1) and cannabis dependence symptoms in adolescents and young adults. *Drug and Alcohol Dependence*, 104(1–2):11–16.

Hasin, D. S., Saha, T. D., Kerridge, B. T., Goldstein, R. B., Chou, S. P., Zhang, H., Jung, J., Pickering, R. P., Ruan, W. J., Smith, S. M., Huang, B., and Grant, B. F. (2015). Prevalence of Marijuana Use Disorders in the United States Between 2001-2002 and 2012-2013. *JAMA psychiatry*, 72(12):1235–1242.

Hawkins, J. D., Catalano, R. F., and Miller, J. Y. (1992). Risk and protective factors for alcohol and other drug problems in adolescence and early adulthood: implications for substance abuse prevention. *Psychological Bulletin*, 112(1):64–105.

Heath, A. C., Bucholz, K. K., Madden, P. A., Dinwiddie, S. H., Slutske, W. S., Bierut, L. J., Statham, D. J., Dunne, M. P., Whitfield, J. B., and Martin, N. G. (1997). Genetic and environmental contributions to alcohol dependence risk in a national twin sample: consistency of findings in women and men. *Psychological Medicine*, 27(6):1381–1396.

Heath, A. C., Jardine, R., and Martin, N. G. (1989). Interactive effects of genotype and social environment on alcohol consumption in female twins. *Journal of Studies on Alcohol*, 50(1):38–48.

Heath, A. C., Kirk, K. M., Meyer, J. M., and Martin, N. G. (1999). Genetic and social determinants of initiation and age at onset of smoking in Australian twins. *Behavior Genetics*, 29(6):395–407.

Heath, A. C., Martin, N. G., Lynskey, M. T., Todorov, A. A., and Madden, P. A. F. (2002). Estimating two-stage models for genetic influences on alcohol, tobacco or drug use initiation and dependence vulnerability in twin and family data. *Twin Research: The Official Journal of the International Society for Twin Studies*, 5(2):113–124.

Hennekens, C. H. and Buring, J. (1987). *Epidemiology in Medicine*. Lippincott, Williams & Wilkins, Philadelphia, PA.

Heron, J., Barker, E. D., Joinson, C., Lewis, G., Hickman, M., Munafo, M., and Macleod, J. (2013). Childhood conduct disorder trajectories, prior risk factors and cannabis use at age 16: birth cohort study. *Addiction*, 108(12):2129–38.

Hesselbrock, M., Easton, C., Bucholz, K. K., Schuckit, M., and Hesselbrock, V. (1999). A validity study of the SSAGA-a comparison with the SCAN. *Addiction*, 94(9):1361–1370.

Hicks, B. M., Durbin, C. E., Blonigen, D. M., Iacono, W. G., and McGue, M. (2012). Relationship between personality change and the onset and course of alcohol dependence in young adulthood. *Addiction*, 107(3):540–8.

Hillstrom, R. P., Cohn, A. M., and Mccarroll, K. A. (1990). Vocal cord paralysis resulting from neck injections in the intravenous drug use population. *The Laryngoscope*, 100(5):503–506.

Hindocha, C., Freeman, T. P., Ferris, J. A., Lynskey, M. T., and Winstock, A. R. (2016). No Smoke without Tobacco: A Global Overview of Cannabis and Tobacco Routes of Administration and Their Association with Intention to Quit. *Addictive Disorders*, page 104.

Hindocha, C., Shaban, N. D. C., Freeman, T. P., Das, R. K., Gale, G., Schafer, G., Falconer, C. J., Morgan, C. J. A., and Curran, H. V. (2015). Associations between cigarette smoking and cannabis dependence: A longitudinal study of young cannabis users in the United Kingdom. *Drug and Alcohol Dependence*, 148:165–171.

Hingson, R. W., Heeren, T., and Edwards, E. M. (2008). Age at Drinking Onset, Alcohol Dependence, and Their Relation to Drug Use and Dependence, Driving Under the Influence of Drugs, and Motor-Vehicle Crash Involvement Because of Drugs. *Journal of Studies on Alcohol and Drugs*, 69(2):192.

Hingson, R. W., Heeren, T., and Winter, M. R. (2006). Age of Alcohol-Dependence Onset: Associations With Severity of Dependence and Seeking Treatment. *Pediatrics*, 118(3):e755–e763.

Hingson, R. W. and Zha, W. (2009). Age of Drinking Onset, Alcohol Use Disorders, Frequent Heavy Drinking, and Unintentionally Injuring Oneself and Others After Drinking. *Pediatrics*, 123(6):1477–1484.

Hopfer, C. (2014). Implications of Marijuana Legalization for Adolescent Substance Use. *Substance Abuse*, 35(4):331–335.

Hopper, J. L. (2002). The Australian Twin Registry. *Twin Research*, 5(5):329–336. WOS:000179240200004.

Hopper, J. L., Foley, D. L., White, P. A., and Pollaers, V. (2013). Australian Twin Registry: 30 years of progress. *Twin Research and Human Genetics: The Official Journal of the International Society for Twin Studies*, 16(1):34–42.

Horyniak, D., Stoové, M., Degenhardt, L., Aitken, C., Kerr, T., and Dietze, P. (2015). How do drug market changes affect characteristics of injecting initiation and subsequent patterns of drug use? findings from a cohort of regular heroin and methamphetamine injectors in melbourne, australia. *Int. J. Drug Policy*, 26.

Hosmer, David W., J., Lemeshow, S., and Sturdivant, R. X. (2013). Model-Building Strategies and Methods for Logistic Regression. In *Applied Logistic Regression*, pages 89–151. John Wiley & Sons, Inc.

Hotopf, M. (2003). The case–control study. In Prince, M., Stewart, R., Ford, T., and Hotopf, M., editors, *Practical Psychiatric Epidemiology*, pages 131–154. Oxford University Press.

Hudziak, J. J., van Beijsterveldt, C. E. M., Bartels, M., Rietveld, M. J. H., Rettew, D. C., Derks, E. M., and Boomsma, D. I. (2003). Individual Differences in Aggression: Genetic Analyses by Age, Gender, and Informant in 3-, 7-, and 10-Year-Old Dutch Twins. *Behavior Genetics*, 33(5):575–589.

Huerta, M., Chodick, G., Balicer, R. D., Davidovitch, N., and Grotto, I. (2005). Reliability of self-reported smoking history and age at initial tobacco use. *Preventive Medicine*, 41(2):646–650. WOS:000230008000036.

Huizink, A. C., Levälahti, E., Korhonen, T., Dick, D. M., Pulkkinen, L., Rose, R. J., and Kaprio, J. (2010). Tobacco, Cannabis, and Other Illicit Drug Use Among Finnish Adolescent Twins: Causal Relationship or Correlated Liabilities? *Journal of Studies on Alcohol and Drugs*, 71(1):5.

Hung, C.-C., Chang, H.-Y., Luh, D.-L., Wu, C.-C., and Yen, L.-L. (2015). Do parents play different roles in drinking behaviours of male and female adolescents? A longitudinal follow-up study. *BMJ Open*, 5(4):e007179.

Hunt, N., Stillwell, G., Taylor, C., and Griffiths, P. (1998). Evaluation of a Brief Intervention to Prevent Initiation into Injecting. *Drugs: Education, Prevention and Policy*, 5(2):185–194.

Jamal, M., Does, A. J., Penninx, B. W. J. H., and Cuijpers, P. (2011). Age at Smoking Onset and the Onset of Depression and Anxiety Disorders. *Nicotine & Tobacco Research*, 13(9):809–819.

Jenkins, S. P. (2008). Preparing survival time data for analysis and estimation.

Johnson, B. A., Cloninger, C. R., Roache, J. D., Bordnick, P. S., and Ruiz, P. (2000). Age of Onset as a Discriminator Between Alcoholic Subtypes in a Treatment-Seeking Outpatient Population. *The American Journal on Addictions*, 9(1):17–27.

Johnson, T. P. and Mott, J. A. (2001). The reliability of self-reported age of onset of tobacco, alcohol and illicit drug use. *Addiction (Abingdon, England)*, 96(8):1187–1198.

Kempel, P., Lampe, K., Parnefjord, R., Hennig, J., and Kunert, H. J. (2003). Auditory-evoked potentials and selective attention: different ways of information processing in cannabis users and controls. *Neuropsychobiology*, 48(2):95–101.

Kendler, K. S. and Baker, J. H. (2007). Genetic influences on measures of the environment: a systematic review. *Psychological Medicine*, 37(5):615–626.

Kendler, K. S. and Gardner, C. O. (1998). Twin studies of adult psychiatric and substance dependence disorders: are they biased by differences in the environmental experiences of monozygotic and dizygotic twins in childhood and adolescence? *Psychological Medicine*, 28(3):625–633.

Kendler, K. S., Jacobson, K. C., Prescott, C. A., and Neale, M. C. (2003). Specificity of Genetic and Environmental Risk Factors for Use and Abuse/Dependence of Cannabis, Cocaine, Hallucinogens, Sedatives, Stimulants, and Opiates in Male Twins. *American Journal of Psychiatry*, 160(4):687–695.

Kendler, K. S., Karkowski, L. M., Corey, L. A., Prescott, C. A., and Neale, M. C. (1999a). Genetic and environmental risk factors in the aetiology of illicit drug initiation and subsequent misuse in women. *The British Journal of Psychiatry*, 175(4):351–356.

Kendler, K. S., Neale, M. C., Kessler, R. C., Heath, A. C., and Eaves, L. J. (1993). A test of the equal-environment assumption in twin studies of psychiatric illness. *Behavior Genetics*, 23(1):21–27.

Kendler, K. S., Neale, M. C., Sullivan, P., Corey, L. A., Gardner, C. O., and Prescott, C. A. (1999b). A population-based twin study in women of smoking initiation and nicotine dependence. *Psychol Med*, 29(2):299–308.

Kendler, K. S., Pedersen, N. L., Farahmand, B. Y., and Persson, P. G. (1996). The treated incidence of psychotic and affective illness in twins compared with population expectation: a study in the Swedish Twin and Psychiatric Registries. *Psychological Medicine*, 26(6):1135–1144.

Kendler, K. S. and Prescott, C. A. (1998). Cannabis Use, Abuse, and Dependence in a Population-Based Sample of Female Twins. *American Journal of Psychiatry*, 155(8):1016–1022.

Kendler, K. S., Schmitt, E., Aggen, S. H., and Prescott, C. A. (2008). Genetic and Environmental Influences on Alcohol, Caffeine, Cannabis, and Nicotine Use From Early Adolescence to Middle Adulthood. *Archives of general psychiatry*, 65(6):674–682.

Kessler, R. C., Berglund, P., Demler, O., Jin, R., Merikangas, K. R., and Walters, E. (2005). LIfetime prevalence and age-of-onset distributions of dsm-iv disorders in the national comorbidity survey replication. *Archives of General Psychiatry*, 62(6):593–602.

Kirkwood, B. R. and Sterne, J. A. C. (2003). *Essential medical statistics*. Blackwell Science, Malden, Mass., 2 edition.

Kleinbaum, D. G. and Klein, M. (2012). *Survival Analysis - A Self-Learning Text, Third Edition*. Springer-Verlag New York.

Laucht, M., Becker, K., Blomeyer, D., and Schmidt, M. H. (2007). Novelty seeking involved in mediating the association between the dopamine D4 receptor gene exon III polymorphism and heavy drinking in male adolescents: results from a high-risk community sample. *Biological Psychiatry*, 61(1):87–92.

Lawrinson, P., Ali, R., Buavirat, A., Chiamwongpaet, S., Dvoryak, S., Habrat, B., Jie, S., Mardiati, R., Mokri, A., Moskalewicz, J., Newcombe, D., Poznyak, V., Subata, E., Uchtenhagen, A., Utami, D. S., Vial, R., and Zhao, C. (2008). Key findings from the WHO collaborative study on substitution therapy for opioid dependence and HIV/AIDS. *Addiction*, 103(9):1484–1492.

Little, R. J. A. and Rubin, D. B. (2014). *Statistical Analysis with Missing Data*. John Wiley & Sons.

Littlefield, A. K., Sher, K. J., and Wood, P. K. (2010). Do changes in drinking motives mediate the relation between personality change and "maturing out" of problem drinking? *Journal of Abnormal Psychology*, 119(1):93–105.

LoParo, D. and Waldman, I. (2014). Twins' Rearing Environment Similarity and Childhood Externalizing Disorders: A Test of the Equal Environments Assumption. *Behavior Genetics*, 44(6):606–613. WOS:000345283700006.

Lopez-Quintero, C., Cobos, J. P. d. l., Hasin, D. S., Okuda, M., Wang, S., Grant, B. F., and Blanco, C. (2011). Probability and predictors of transition from first use to dependence on nicotine, alcohol, cannabis, and cocaine: Results of the National Epidemiologic Survey on Alcohol and Related Conditions (NESARC). *Drug and Alcohol Dependence*, 115(1–2):120–130.

Lynskey, M. and Agrawal, A. (2007). Psychometric properties of dsm assessments of illicit drug abuse and dependence: results from the national epidemiologic survey on alcohol and related conditions (nesarc. *Psychological Medicine*, 9:1345–1355.

Lynskey, M. T., Agrawal, A., and Heath, A. C. (2010). Genetically informative research on adolescent substance use: methods, findings and challenges. *Journal of the American Academy of Child and Adolescent Psychiatry*, 49(12):1202–1214.

Lynskey, M. T., Agrawal, A., Henders, A., Nelson, E. C., Madden, P. A. F., and Martin, N. G. (2012). An Australian Twin Study of Cannabis and Other Illicit Drug Use and Misuse, and Other Psychopathology. *Twin Research and Human Genetics*, 15(05):631–641.

Lynskey, M. T., Fergusson, D. M., and Horwood, L. J. (1998). The Origins of the Correlations between Tobacco, Alcohol, and Cannabis Use During Adolescence. *Journal of Child Psychology and Psychiatry*, 39(7):995–1005.

Lynskey, M. T. and Hall, W. (1998). Cohort trends in age of initiation to heroin use. *Drug and Alcohol Review*, 17(3):289–297.

Lynskey, M. T., Heath, A. C., Bucholz, K. K., Slutske, W. S., Madden, P. A., Nelson, E. C., Statham, D. J., and Martin, N. G. (2003). Escalation of drug use in early-onset cannabis users vs co-twin controls. *JAMA*, 289(4):427–33.

Lynskey, M. T., Heath, A. C., Nelson, E. C., Bucholz, K. K., Madden, P. A. F., Slutske, W. S., Statham, D. J., and Martin, N. G. (2002). Genetic and environmental contributions to cannabis dependence in a national young adult twin sample. *Psychological Medicine*, 32(02):195–207.

Lynskey, M. T., Vink, J. M., and Boomsma, D. I. (2006). Early Onset Cannabis Use and Progression to other Drug Use in a Sample of Dutch Twins. *Behavior Genetics*, 36(2):195–200.

Lytton, H. (1977). Do parents create, or respond to, differences in twins? *Developmental Psychology*, 13(5):456–459.

Maas, C. J. M. and Hox, J. J. (2004). Robustness issues in multilevel regression analysis. *Statistica Neerlandica*, 58(2):127–137.

Maes, H. H., Sullivan, P. F., Bulik, C. M., Neale, M. C., Prescott, C. A., Eaves, L. J., and Kendler, K. S. (2004a). A twin study of genetic and environmental influences on tobacco initiation, regular tobacco use and nicotine dependence. *Psychological Medicine*, 34(7):1251–1261. WOS:000225508200011.

Maes, H. H., Sullivan, P. F., Bulik, C. M., Neale, M. C., Prescott, C. A., Eaves, L. J., and Kendler, K. S. (2004b). A twin study of genetic and environmental influences on tobacco initiation, regular tobacco use and nicotine dependence. *Psychological Medicine*, 34(7):1251–1261.

Maliphant, J. and Scott, J. (2005). Use of the femoral vein ('groin injecting') by a sample of needle exchange clients in Bristol, UK. *Harm Reduction Journal*, 2:6.

Martins, S. S., Sampson, L., Cerdá, M., and Galea, S. (2015). Worldwide Prevalence and Trends in Unintentional Drug Overdose: A Systematic Review of the Literature. *American Journal of Public Health*, 105(11):2373–2373.

McCutcheon, V. V., Grant, J. D., Heath, A. C., Bucholz, K. K., Sartor, C. E., Nelson, E. C., Madden, P. a. F., and Martin, N. G. (2012). Environmental influences predominate in remission from alcohol use disorder in young adult twins. *Psychological Medicine*, 42(11):2421–2431.

McGeary, J. E., Knopik, V. S., Hayes, J. E., Palmer, R. H., Monti, P. M., and Kalman, D. (2012). Predictors of relapse in a bupropion trial for smoking cessation in recently-abstinent alcoholics: preliminary results using an aggregate genetic risk score. *Substance Abuse: Research and Treatment*, 6:107–114.

McGue, M. and Bouchard, T. J. (1984). Adjustment of twin data for the effects of age and sex. *Behavior Genetics*, 14(4):325–343.

Merikangas KR, Stolar M, Stevens DE, and et al (1998). Familial transmission of substance use disorders. *Archives of General Psychiatry*, 55(11):973–979.

Miles, D. R., Stallings, M. C., Young, S. E., Hewitt, J. K., Crowley, T. J., and Fulker, D. W. (1998). A family history and direct interview study of the familial aggregation of substance abuse: the adolescent substance abuse study. *Drug and Alcohol Dependence*, 49(2):105–114.

Miller, J. E., Nugent, C. N., and Russell, L. B. (2015). Risk factors for family time burdens providing and arranging health care for children with special health care needs: Lessons from nonproportional odds models. *Social Science Research*, 52:602–614.

Miller, P. G., Hyder, S., Zinkiewicz, L., Droste, N., and Harris, J. B. (2014). Comparing subjective well-being and health-related quality of life of Australian drug users in treatment in Regional and Rural Victoria. *Drug and Alcohol Review*, 33(6):651–657.

Mills, K. L., Teesson, M., Darke, S., Ross, J., and Lynskey, M. (2004). Young people with heroin dependence: Findings from the Australian Treatment Outcome Study (ATOS). *Journal of Substance Abuse Treatment*, 27(1):67–73.

Moffitt, T. E. (1993). Adolescence-limited and life-course-persistent antisocial behavior: a developmental taxonomy. *Psychological Review*, 100:674–701.

Moffitt, T. E., Caspi, A., and Rutter, M. (2005). Strategy for investigating interactions between measured genes and measured environments. *Archives of General Psychiatry*, 62(5):473–481.

Morley, K. I., Lynskey, M. T., Madden, P. A. F., Treloar, S. A., Heath, A. C., and Martin, N. G. (2007). Exploring the inter-relationship of smoking age-at-onset, cigarette consumption and smoking persistence: genes or environment? *Psychological Medicine*, 37(9):1357–1367. WOS:000249071900015.

Morral, A. R., McCaffrey, D. F., and Paddock, S. M. (2002). Reassessing the marijuana gateway effect. *Addiction (Abingdon, England)*, 97(12):1493–1504.

Neale, M. and Cardon, L. (1992). *Methodology for genetic studies of twins and families*. Kluwer Academic Publications, Dordrecht, The Netherlands.

Neale, M. C., Aggen, S. H., Maes, H. H., Kubarych, T. S., and Schmitt, J. E. (2006a). Methodological issues in the assessment of substance use phenotypes. *Addictive Behaviors*, 31(6):1010–1034.

Neale, M. C., Eaves, L. J., and Kendler, K. S. (1994). The power of the classical twin study to resolve variation in threshold traits. *Behavior Genetics*, 24(3):239–258.

Neale, M. C., Harvey, E., Maes, H. H., Sullivan, P. F., and Kendler, K. S. (2006b). Extensions to the modeling of initiation and progression: applications to substance use and abuse. *Behav Genet*, 36(4):507–24.

Neale, M. C., Røysamb, E., and Jacobson, K. (2006c). Multivariate Genetic Analysis of Sex Limitation and G × E Interaction. *Twin research and human genetics : the official journal of the International Society for Twin Studies*, 9(4):481–489.

Nelson, E. C., Lynskey, M. T., Heath, A. C., Wray, N., Agrawal, A., Shand, F. L., Henders, A. K., Wallace, L., Todorov, A. A., Schrage, A. J., Saccone, N. L., Madden, P. A., Degenhardt, L., Martin, N. G., and Montgomery, G. W. (2013). ANKK1, TTC12, and NCAM1 polymorphisms and heroin dependence: importance of considering drug exposure. *JAMA Psychiatry*, 70(3):325–33.

Newton, N. C., Conrod, P. J., Slade, T., Carragher, N., Champion, K. E., Barrett, E. L., Kelly, E. V., Nair, N. K., Stapinski, L., and Teesson, M. (2016). The long-term effectiveness of a selective, personality-targeted prevention program in reducing alcohol use and related harms: a cluster randomized controlled trial. *Journal of Child Psychology and Psychiatry, and Allied Disciplines*, 57(9):1056–1065.

of Health and Welfare, A. I. (2014). National Drug Strategy Household Survey detailed report: 2013. Technical report, AIHW, Canberra.

O'Keefe, D., Horyniak, D., and Dietze, P. (2016). From initiating injecting drug use to regular injecting: Retrospective survival analysis of injecting progression within a sample of people who inject drugs regularly. *Drug and Alcohol Dependence*, 158:177–180.

Ottena, R., Barkerb, E. D., Maughanc, B., Arseneault, L., and Engelsa, R. C. (2010). Self-control and its relation to joint developmental trajectories of cannabis use and depressive mood symptoms. *Drug and Alcohol Dependence*, 112(3):201–208.

Pacek, L. R., Martins, S. S., and Crum, R. M. (2013). The bidirectional relationships between alcohol, cannabis, co-occurring alcohol and cannabis use disorders with major depressive disorder: results from a national sample. *Journal of Affective Disorders*, 148(2-3):188–195.

Pacula, R. L., Kilmer, B., Wagenaar, A. C., Chaloupka, F. J., and Caulkins, J. P. (2014). Developing public health regulations for marijuana: Lessons from alcohol and tobacco. *American Journal of Public Health*, 104(6):1021–1028.

Pagan, J. L., Rose, R. J., Viken, R. J., Pulkkinen, L., Kaprio, J., and Dick, D. M. (2006). Genetic and Environmental Influences on Stages of Alcohol Use Across Adolescence and into Young Adulthood. *Behavior Genetics*, 36(4):483–497.

Palmer, R. H. C., Button, T. M., Rhee, S. H., Corley, R. P., Young, S. E., Stallings, M. C., Hopfer, C. J., and Hewitt, J. K. (2012). Genetic etiology of the common liability to drug dependence: evidence of common and specific mechanisms for DSM-IV dependence symptoms. *Drug and Alcohol Dependence*, 123 Suppl 1:S24–32.

Parra, G. R., O'Neill, S. E., and Sher, K. J. (2003). Reliability of self-reported age of substance involvement onset. *Psychology of Addictive Behaviors*, 17(3):211–218.

Patton, G. C., Coffey, C., Carlin, J. B., Degenhardt, L., Lynskey, M., and Hall, W. (2002). Cannabis use and mental health in young people: cohort study. *BMJ*, 325(7374):1195–1198.

Pedersen, W., Mastekaasa, A., and Wichstrøm, L. (2001). Conduct problems and early cannabis initiation: a longitudinal study of gender differences. *Addiction*, 96(3):415–431.

Pinchevsky, G. M., Arria, A. M., Caldeira, K. M., Garnier-Dykstra, L. M., Vincent, K. B., and O'Grady, K. E. (2012). Marijuana Exposure Opportunity and Initiation during College: Parent and Peer Influences. *Prevention Science*, 13(1):43–54.

Pitkänen, T., Lyyra, A.-L., and Pulkkinen, L. (2005). Age of onset of drinking and the use of alcohol in adulthood: a follow-up study from age 8–42 for females and males. *Addiction*, 100(5):652–661.

Pizzey, R. and Hunt, N. (2008). Distributing foil from needle and syringe programmes (NSPs) to promote transitions from heroin injecting to chasing: an evaluation. *Harm Reduction Journal*, 5:24.

Plomin, R., DeFries, J. C., Knopik, V. S., and Neiderheiser, J. (2013). *Behavioral Genetics*. Worth Publishers, New York, 6 edition.

Plomin, R., Willerman, L., and Loehlin, J. C. (1976). Resemblance in appearance and the equal environments assumption in twin studies of personality traits. *Behavior Genetics*, 6(1):43–52.

Posthuma, D., Beem, A. L., de Geus, E. J. C., van Baal, G. C. M., von Hjelmborg, J. B., Iachine, I., and Boomsma, D. I. (2003). Theory and Practice in Quantitative Genetics. *Twin Research*, 6(5):361–376.

Poznyak, V. B., White, J., and Clark, N. (2011). *Substance Abuse Disorders: Evidence and Experience*, chapter Drug Abuse: Prevention. John Wiley and Sons.

Prince, M. (2003). Cross-sectional surveys. In Prince, M., Stewart, R., Ford, T., and Hotopf, M., editors, *Practical Psychiatric Epidemiology*, pages 111–129. Oxford University Press.

R Core Team (2013). R: A language and environment for statistical computing.

Rang, H. P. and Dale, M. M., editors (2016). *Rang and Dale's pharmacology*. Elsevier/Churchill Livingstone, Edinburgh, 8th ed edition.

Reboussin, B. A., Hubbard, S., and Ialongo, N. S. (2007). Marijuana Use Patterns among African-American Middle-School Students. *Drug and alcohol dependence*, 90(1):12–24.

Reboussin, B. A., Ialongo, N. S., and Green, K. M. (2015). Influences of behavior and academic problems at school entry on marijuana use transitions during adolescence in an African American sample. *Addictive behaviors*, 41:51–57.

Reef, J., Diamantopoulou, S., Meurs, I. v., Verhulst, F. C., and Ende, J. v. d. (2010). Developmental trajectories of child to adolescent externalizing behavior and adult DSM-IV disorder: results of a 24-year longitudinal study. *Social Psychiatry and Psychiatric Epidemiology*, 46(12):1233–1241.

Reich, T., Hinrichs, A., Culverhouse, R., , and Bierut, L. (1999). Genetic studies of alcoholism and substance dependencependence. *Psychiatric Genetics*, 65:599–605.

Rice, J. P., Reich, T., Bucholz, K. K., Neuman, R. J., Fishman, R., Rochberg, N., Hesselbrock, V. M., Nurnberger, J. I., Schuckit, M. A., and Begleiter, H. (1995). Comparison of direct interview and family history diagnoses of alcohol dependence. *Alcoholism, Clinical and Experimental Research*, 19(4):1018–1023.

Ridenour, T. A., Lanza, S. T., Donny, E. C., and Clark, D. B. (2006). Different lengths of times for progressions in adolescent substance involvement. *Addictive behaviors*, 31(6):962–983.

Ridenour, T. A., Maldonado-Molina, M., Compton, W. M., Spitznagel, E. L., and Cottler, L. B. (2005). Factors associated with the transition from abuse to dependence among substance abusers: Implications for a measure of addictive liability. *Drug and alcohol dependence*, 80(1):1–14.

Rifai, N., Gillette, M. A., and Carr, S. A. (2006). Protein biomarker discovery and validation: the long and uncertain path to clinical utility. *Nature Biotechnology*, 24(8):971–983.

Ropponen, A. and Svedberg, P. (2014). Single and additive effects of health behaviours on the risk for disability pensions among Swedish twins. *The European Journal of Public Health*, 24(4):643–648.

Ross, J., Teeson, M., Darke, S., Lynskey, M., Hetherington, K., Mills, K., Williamson, S., and Fairburn, S. (2003). Characteristics of heroin users entering three treatment modalities in New South Wales: Baseline findings from the Australian Treatment Outcome Study (ATOS). Technical Report 139, NDARC.

Salvatore, J. E., Aliev, F., Edwards, A. C., Evans, D. M., Macleod, J., Hickman, M., Lewis, G., Kendler, K. S., Loukola, A., Korhonen, T., Latvala, A., Rose, R. J., Kaprio, J., and Dick, D. M. (2014). Polygenic scores predict alcohol problems in an independent sample and show moderation by the environment. *Genes*, 5(2):330–346.

Sartor, C. E., Agrawal, A., Grant, J. D., Duncan, A. E., Madden, P. A. F., Lynskey, M. T., Heath, A. C., and Bucholz, K. K. (2015). Differences Between African-American and European- American Women in the Association of Childhood Sexual Abuse With Initiation of Marijuana Use and Progression to Problem Use. *Journal of Studies on Alcohol and Drugs*, 76(4):569–577.

Sartor, C. E., Agrawal, A., Lynskey, M. T., Bucholz, K. K., and Heath, A. C. (2008). Genetic and environmental influences on the rate of progression to alcohol dependence in young women. *Alcoholism, clinical and experimental research*, 32(4):632–8.

Sartor, C. E., Agrawal, A., Lynskey, M. T., Bucholz, K. K., Madden, P. A., and Heath, A. C. (2009a). Common genetic influences on the timing of first use for alcohol, cigarettes, and cannabis in young African-American women. *Drug and alcohol dependence*, 102(1-3):49–55.

Sartor, C. E., Agrawal, A., Lynskey, M. T., Duncan, A. E., Grant, J. D., Nelson, E. C., Madden, P. a. F., Heath, A. C., and Bucholz, K. K. (2013a). Cannabis or alcohol first? Differences by ethnicity and in risk for rapid progression to cannabis-related problems in women. *Psychological Medicine*, 43(4):813–823.

Sartor, C. E., Grant, J. D., Bucholz, K. K., Madden, P. A. F., Heath, A. C., Agrawal, A., Whitfield, J. B., Statham, D. J., Martin, N. G., and Lynskey, M. T. (2010).

Common Genetic Contributions to Alcohol and Cannabis Use and Dependence Symptomatology. *Alcoholism-Clinical and Experimental Research*, 34(3):545–554. WOS:000275142100019.

Sartor, C. E., Lynskey, M. T., Bucholz, K. K., Madden, P. A. F., Martin, N. G., and Heath, A. C. (2009b). Timing of first alcohol use and alcohol dependence: evidence of common genetic influences. *Addiction*, 104(9):1512–1518. WOS:000268653300013.

Sartor, C. E., Lynskey, M. T., Heath, A. C., Jacob, T., and True, W. (2007). The role of childhood risk factors in initiation of alcohol use and progression to alcohol dependence. *Addiction*, 102(2):216–225. WOS:000243943000012.

Sartor, C. E., Waldron, M., Duncan, A. E., Grant, J. D., McCutcheon, V. V., Nelson, E. C., Madden, P. A., Bucholz, K. K., and Heath, A. C. (2013b). Childhood Sexual Abuse and Early Substance Use in Adolescent Girls: The Role of Familial Influences. *Addiction (Abingdon, England)*, 108(5):993–1000.

SAS Institute Inc (2002). SAS Software version 9.3.

Schemper, M. (1992). Cox Analysis of Survival Data with Non-Proportional Hazard Functions. *Journal of the Royal Statistical Society. Series D (The Statistician)*, 41(4):455–465.

Schulenberg, J. E. and Maggs, J. L. (2002). A developmental perspective on alcohol use and heavy drinking during adolescence and the transition to young adulthood. *Journal of Studies on Alcohol, Supplement*, (s14):54–70.

Sher, K. J., Dick, D. M., Crabbe, J. C., Hutchison, K. E., O'Malley, S. S., and Heath, A. C. (2010). Consilient research approaches in studying gene × environment interactions in alcohol research. *Addiction Biology*, 15(2):200–216.

Sherva, R., Wang, Q., Kranzler, H., Zhao, H., Koesterer, R., Herman, A., Farrer, L., and Gelernter, J. (2016). GEnome-wide association study of cannabis dependence severity, novel risk variants, and shared genetic risks. *JAMA Psychiatry*, 73(5):472–480.

Shi, Y., Lenzi, M., and An, R. (2015). Cannabis Liberalization and Adolescent Cannabis Use: A Cross-National Study in 38 Countries. *PloS One*, 10(11):e0143562.

Shillington, A. M., Cottler, L. B., Mager, D. E., and Compton III, W. M. (1995). Self-report stability for substance use over 10 years: data from the St. Louis Epidemiologic Catchment Study. *Drug and Alcohol Dependence*, 40(2):103–109.

Singer, E. and Presser, S. (2007). Privacy, Confidentiality, and Respondent Burden as Factors in Telephone Survey Nonresponse. In Lepkowski, J. M., Tucker, C., Brick, J. M., Leeuw, E. D. d., Japec, L., Lavrakas, P. J., Link, M. W., and Sangster, R. L., editors, *Advances in Telephone Survey Methodology*, pages 447–470. John Wiley & Sons, Inc.

Skosnik, P. D., Krishnan, G. P., Vohs, J. L., and O'Donnell, B. F. (2006). The effect of cannabis use and gender on the visual steady state evoked potential. *Clinical Neurophysiology: Official Journal of the International Federation of Clinical Neurophysiology*, 117(1):144–156.

Stallings, M. C., Hewitt, J. K., Beresford, T., Heath, A. C., and Eaves, L. J. (1999). A Twin Study of Drinking and Smoking Onset and Latencies from First Use to Regular Use. *Behavior Genetics*, 29(6):409–421.

StataCorp (2009). Stata Statistical Software: Release 11.

Stockings, E., Hall, W. D., Lynskey, M., Morley, K. I., Reavley, N., Strang, J., Patton, G., and Degenhardt, L. (2016). Prevention, early intervention, harm reduction, and treatment of substance use in young people. *The Lancet Psychiatry*, 3(3):280–296.

Stoltman, J. J. K., Woodcock, E. A., Lister, J. J., Greenwald, M. K., and Lundahl, L. H. (2015). Exploration of the telescoping effect among not-in-treatment, intensive heroin-using research volunteers. *Drug and Alcohol Dependence*, 148:217–220.

Storr, C. L., Wagner, F. A., Chen, C. Y., and Anthony, J. C. (2011). Childhood predictors of first chance to use and use of cannabis by young adulthood. *Drug and alcohol dependence*, 117(1):7–15.

Strang, J. (1988). Changing Injecting Practices: blunting the needle habit. *British Journal of Addiction*, 83(3):237–239.

Strang, J., Bearn, J., Farrell, M., Finch, E., Gossop, M., Griffiths, P., Marsden, J., and Wolff, K. (1998). Route of drug use and its implications for drug effect, risk of dependence and health consequences. *Drug and Alcohol Review*, 17(2):197–211.

Strang, J., Griffiths, P., Powis, B., and Gossop, M. (1992). First use of heroin: changes in route of administration over time. *BMJ (Clinical research ed.)*, 304(6836):1222–1223.

Strang, J. and King, L. (1997). Heroin is more than just diamorphine. *Addiction Research*, 5(1).

Strang, J. and McCambridge, J. (2005). Are cannabis users exposed to other drug use opportunities? Investigation of high-risk drug exposure opportunities among young cannabis users in London. *Drug and Alcohol Review*, 24(2):185–191.

Sun, G. W., Shook, T. L., and Kay, G. L. (1996). Inappropriate use of bivariable analysis to screen risk factors for use in multivariable analysis. *Journal of Clinical Epidemiology*, 49(8):907–916.

Swendsen, J., Anthony, J. C., Conway, K. P., Degenhardt, L., Dierker, L., Glantz, M., He, J., Kalaydjian, A., Kessler, R. C., Sampson, N., and Merikangas, K. R. (2008). Improving targets for the prevention of drug use disorders: Sociodemographic predictors of transitions across drug use stages in the national comorbidity survey replication. *Preventive Medicine*, 47(6):629–634.

Swift, W., Coffey, C., Carlin, J. B., Degenhardt, L., and Patton, G. C. (2008). Adolescent cannabis users at 24 years: trajectories to regular weekly use and dependence in young adulthood. *Addiction*, 103(8):1361–70.

Tabor, H. K., Risch, N. J., and Myers, R. M. (2002). Candidate-gene approaches for studying complex genetic traits: practical considerations. *Nature Reviews Genetics*, 3:391–397.

Tas, B. and Day, E. (2016). Pharmacology and physiological mechanisms of opioid overdose and reversal. In *Preventing opioid overdose deaths with take-home*

naloxone, number 20 in Insights. EMCDDA, European Monitoring Centre for Drugs and Drug Addiction, Lisbon.

Teesson, M., Newton, N. C., Slade, T., Chapman, C., Allsop, S., Hides, L., McBride, N., Mewton, L., Tonks, Z., Birrell, L., Brownhill, L., and Andrews, G. (2014). The CLIMATE schools combined study: a cluster randomised controlled trial of a universal Internet-based prevention program for youth substance misuse, depression and anxiety. *BMC psychiatry*, 14:32.

Tsuang, M. T., Lyons, M. J., Harley, R. M., Xian, H., Eisen, S., Goldberg, J., True, W. R., and Faraone, S. V. (1999). Genetic and environmental influences on transitions in drug use. *Behav Genet*, 29(6):473–9.

Tsuang, M. T., Lyons, M. J., Meyer, J. M., Doyle, T., Eisen, S. A., Goldberg, J., True, W., Lin, N., Toomey, R., and Eaves, L. (1998). Co-occurrence of abuse of different drugs in men: the role of drug-specific and shared vulnerabilities. *Archives of General Psychiatry*, 55(11):967–972.

UNODC (2014). World Drug Report 2014. United Nations publication, Sales No. E.12.XI.1), United Nations publication.

Valdés, E. G., Andel, R., Sieurin, J., Feldman, A. L., Edwards, J. D., Långström, N., Gatz, M., and Wirdefeldt, K. (2014). Occupational Complexity and Risk of Parkinson's Disease. *PLoS ONE*, 9(9).

van der Zwaluw, C. S., Otten, R., Kleinjan, M., and Engels, R. C. M. E. (2014). Different trajectories of adolescent alcohol use: testing gene-environment interactions. *Alcoholism, Clinical and Experimental Research*, 38(3):704–712.

van Etten, M. L. and Anthony, J. C. (1999). Comparative epidemiology of initial drug opportunities and transitions to first use: marijuana, cocaine, hallucinogens and heroin. *Drug and Alcohol Dependence*, 54(2):117–125.

van Etten, M. L., Neumark, Y., and Anthony, J. C. (1997). Initial opportunity to use marijuana and the transition to first use: United states, 1979–1994. *Drug and Alcohol Dependence*, 49:1–7.

van Etten, M. L., Neumark, Y. D., and Anthony, J. C. (1999). Male-female differences in the earliest stages of drug involvement. *Addiction*, 94(9):1413–1419.

Verweij, K. J. H., Creemers, H. E., Korhonen, T., Latvala, A., Dick, D. M., Rose, R. J., Huizink, A. C., and Kaprio, J. (2016). Role of overlapping genetic and environmental factors in the relationship between early adolescent conduct problems and substance use in young adulthood. *Addiction*, 111(6):1036–1045.

Verweij, K. J. H., Mosing, M. A., Zietsch, B. P., and Medland, S. E. (2012). Estimating Heritability from Twin Studies. In Elston, R. C., Satagopan, J. M., and Sun, S., editors, *Statistical Human Genetics*, number 850 in Methods in Molecular Biology, pages 151–170. Humana Press.

Verweij, K. J. H., Zietsch, B. P., Lynskey, M. T., Medland, S. E., Neale, M. C., Martin, N. G., Boomsma, D. I., and Vink, J. M. (2010). Genetic and environmental influences on cannabis use initiation and problematic use: a meta-analysis of twin studies. *Addiction*, 105(3):417–430.

Vitaro, F., Brendgen, M., and Arseneault, L. (2009). The discordant MZ-twin method: One step closer to the holy grail of causality. *International Journal of Behavioral Development*, 33(4):376–382. WOS:000267323600011.

Vittinghoff, E., Glidden, D. V., Shiboski, S. C., and McCulloch, C. E. (2005). *Regression Methods in Biostatistics*. Statistics for Biology and Health. Springer, New York.

Vogt, W. P. and Johnson, R. B. (2016). *Dictionary of Statistics & Methodology: A Nontechnical Guide for the Social Sciences*. Sage Publications, 5 edition.

Wagner, F. A. and Anthony, J. C. (2002). Into the world of illegal drug use: Exposure opportunity and other mechanisms linking the use of alcohol, tobacco, marijuana, and cocaine. *Journal of Epidemiology*, pages 918–925.

Wagner, F. A. and Anthony, J. C. (2007). Male–female differences in the risk of progression from first use to dependence upon cannabis, cocaine, and alcohol. *Drug and Alcohol Dependence*, 86(2–3):191–198.

Waldron, M., Grant, J. D., Bucholz, K. K., Lynskey, M. T., Slutske, W. S., Glowinski, A. L., Henders, A., Statham, D. J., Martin, N. G., and Heath, A. C. (2014). Parental separation and early substance involvement: Results from children of alcoholic and cannabis dependent twins. *Drug and Alcohol Dependence*, 134:78–84.

Warner-Smith, M., Darke, S., Lynskey, M., and Hall, W. (2001). Heroin overdose: causes and consequences. *Addiction (Abingdon, England)*, 96(8):1113–1125.

Weiss, R. D., Griffin, M. L., and Mirin, S. M. (1992). Drug Abuse as Self-Medication for Depression: An Empirical Study. *The American Journal of Drug and Alcohol Abuse*, 18(2):121–129.

West, R. and Brown, J. (2013). Addiction, habit and instrumental learning. In *Theory of Addiction*, pages 114–135. John Wiley & Sons, Ltd.

Williams, J. and Hagger-Johnson, G. (2017). Childhood academic ability in relation to cigarette, alcohol and cannabis use from adolescence into early adulthood: Longitudinal study of young people in england (lsype). *BMJ Open*, 7(2).

Williams, R. (2006). Generalized ordered logit/partial proportional odds models for ordinal dependent variables. *Stata Journal*, 6(1):58–82.

Winters, K. C. and Lee, C.-Y. S. (2008). Likelihood of developing an alcohol and cannabis use disorder during youth: Association with recent use and age. *Drug and Alcohol Dependence*, 92(1–3):239–247.

Wittchen, H.-U., Behrendt, S., Höfler, M., Perkonigg, A., Lieb, R., Bühringer, G., and Beesdo, K. (2008). What are the high risk periods for incident substance use and transitions to abuse and dependence? Implications for early intervention and prevention. *International Journal of Methods in Psychiatric Research*, 17(S1):S16–S29.

Wodak, A. and McLeod, L. (2008). The role of harm reduction in controlling hiv among injecting drug users. *AIDS*, 22:81–92.

Woodcock, E. A., Lundahl, L. H., Stoltman, J. J. K., and Greenwald, M. K. (2015). Progression to regular heroin use: Examination of patterns, predictors, and consequences. *Addictive Behaviors*, 45:287–293.

Wu, P., Bird, H. R., Liu, X., Fan, B., Fuller, C., Shen, S., Duarte, C. S., and Canino, G. J. (2006). Childhood Depressive Symptoms and Early Onset of Alcohol Use. *Pediatrics*, 118(5):1907–1915.

Wulf-Johansson, H., Thinggaard, M., Tan, Q., Johansson, S. L., Schlosser, A., Christensen, K., Holmskov, U., and Sorensen, G. L. (2013). Circulating surfactant protein D is associated to mortality in elderly women: A twin study. *Immunobiology*, 218(5):712–717.

Young, S. E., Corley, R. P., Stallings, M. C., Rhee, S. H., Crowley, T. J., and Hewitt, J. K. (2002). Substance use, abuse and dependence in adolescence: prevalence, symptom profiles and correlates. *Drug and Alcohol Dependence*, 68(3):309–322.

Zohsel, K., Baldus, C., Schmidt, M. H., Esser, G., Banaschewski, T., Thomasius, R., and Laucht, M. (2016). Predicting later problematic cannabis use from psychopathological symptoms during childhood and adolescence: Results of a 25-year longitudinal study. *Drug and Alcohol Dependence*, 163:251–255. WOS:000378457000034.

.1 Appendix 1: Quality Rating Table

Guide to quality ratings assigned to papers identified through the systematic literature search (Chapter 1).

Criteria	Score 0	Score 1	Score 2	Score 3
Type of Study	-	Observations without control	Cohort or case control	-
Selection Method	Non-random/ Convenience sample	Record review/ Clinic sample	Random/ Representative Sample	-
Study Aim	Unrelated to transition speed	Aimed to study transition	-	-
Prospective	No	Yes	-	-
Control Group	No control	Non-Users/ Had not reached stage	Slower/Faster transitioners	-
Measure of Speed	By specified point e.g. "early"	Continuous age at stage	Time from one stage to next	-
Analysis Method	Other	Linear/Logistic regresison	Survival Analysis/Discrete Time Hazard Model	-
Sample Size	$\leq$500	500 - 1000	1000 - 5000	$\geq$5000
Response Rate	No/NA	Yes	-	-

.2 Appendix 2: Missing Data

Table of all analysis variables for each sample, and missing data for each variable (starts on next page).

Category	Variable	Australian Twin Registry Sample		SLAM Opiate Treatment Sample		1990s Drug Transitions Study Sample	
		Included in anlayses ✓or ×	Non-structural missing data N	Included in anlayses ✓or ×	Non-Structural missing data N	Included in anlayses ✓or ×	Non-Structural missing data N
Cannabis use	Lifetime opportunity to use	✓	26	×	-	×	-
	Age of opportunity onset	✓	0	×	-	×	-
	Lifetime use	✓	27	×	-	×	-
	Age of initiaiton	✓	1	×	-	×	-
	Dependence	✓	0	×	-	×	-
	Age of dependence onset	✓	0	×	-	×	-
Heroin use	Lifetime opportunity to use	×	-	✓	0	×	-
	Age of opportunity onset	×	-	✓	3	×	-
	Lifetime use	×	-	✓	0	✓	0
	Age of initiaiton onset	×	-	✓	0	✓	0
	Year of initiaiton	×	-	×	-	✓	7
	Time from opportunity to initiation	×	-	✓	5	×	-
	Route of iniital administration	×	-	×	-	✓	5
	Subsequent use	×	-	✓	0	×	-
	Time from initiation to subsequent use	×	-	✓	0	×	-
	Daily use	×	-	✓	2	×	-
	Age of daily use onset	×	-	✓	2	×	-
	Time from initiation to daily use	×	-	×	-	✓	12
	Problematic use	×	-	✓	0	×	-
	Age of problem onset	×	-	✓	0	×	-
	Heavy opiate use	×	-	✓	5	×	-
	Injecting into groin/neck	×	-	✓	-	×	-
	Lifetime overdose	×	-	✓	0	×	-
	Age at treatment seeking	×	-	✓	2	×	-
	Severity of dependence at treatment entry	×	-	✓	0	×	-
	In treatment at interview	×	-	×	-	✓	0

Table continued on next page

Category	Variable	Australian Twin Registry Sample		SLAM Opiate Treatment Sample		1990s Drug Transitions Study Sample	
		Included in anlayses ✓or ×	Non-structural missing data N	Included in anlayses ✓or ×	Non-Structural missing data N	Included in anlayses ✓or ×	Non-Structural missing data N
Demographic data	Age at interview	✓	0	✓	0	×	-
	Gender	✓	0	✓	1	✓	0
	Level of completed education	✓	3	✓	0	×	-
	Ethnicity	×	-	✓	0	✓	11
Parenting factors	Strict parenting	✓	6	×	-	×	-
	Alcohol problems	✓	2	×	-	×	-
	Drug problems	✓	2	×	-	×	-
	Single parent family	✓	2	×	-	×	-
	High parent conflict	✓	2	×	-	×	-
Childhood Factors	Religious attendance	✓	2	×	-	×	-
	Childhood sexual abuse (CSA)	✓	43	×	-	×	-
	CSA age of onset	✓	4	×	-	×	-
	Peer cannabis use	✓		×	-	×	-
Mental health	Conduct disorder (CD)	✓	0	×	-	×	-
	Age at CD onset	✓	0	×	-	×	-
	Depressive episode	✓	36	×	-	×	-
	Age at depressive episode onset	✓	7	×	-	×	-

Table continued on next page

Category	Variable	Australian Twin Registry Sample		SLAM Opiate Treatment Sample		1990s Drug Transitions Study Sample	
		Included in anlayses ✓or ×	Non-structural missing data N	Included in anlayses ✓or ×	Non-Structural missing data N	Included in anlayses ✓or ×	Non-Structural missing data N
Tobacco, alcohol and other drug use	Lifetime tobacco use	✓	3	×	-	×	-
	Age at tobacco initiation	✓	2	×	-	×	-
	Lifetime weekly tobacco use	✓	4	×	-	×	-
	Age at weekly tobacco use onset	✓	1	×	-	×	-
	Lifetime tobacco dependence	✓	0	×	-	×	-
	Age at tobacco dependence onset	✓	1	×	-	×	-
	Lifetime alcohol use	✓	13	×	-	×	-
	Age at alcohol initiation	✓	2	×	-	×	-
	Lifetime monthly alcohol use	✓	14	×	-	×	-
	Age at monthly alcohol use onset	✓	0	×	-	×	-
	Lifetime alcohol dependence	✓	0	×	-	×	-
	Age at alcohol dependence onset	✓	0	×	-	×	-
	Lifetime other drug use	✓	0	×	-	×	-
	Age at other drug initiation	✓	2	×	-	×	-
	Lifetime other drug dependence	✓	0	×	-	×	-
	Age at other drug dependence onset	✓	0	×	-	×	-
	Regular use of other drugs before heroin initiation	×	-	×	-	✓	5

.3 Appendix 3: Association Between ATR Analysis Outcomes and Study Covariates

Table of OR (95% CI) for the associaiton between model covariates and cannabis outcomes included in analyses in Chapters 3 and 4.

Covariate	Daily cannabis use		Cannabis Abuse and/or Dependence		Cannabis Treatment Seeking	
	OR (95 % CI)	P	OR (95 % CI)	P	OR (95 % CI)	P
Gender - male	**1.86** (1.38-2.51)	≤0.001	**2.22** (1.74-2.84)	≤0.001	**1.74** (1.11-2.74)	0.02
Lower level of completed education	**1.58** (1.19-2.10)	0.002	**1.28** (1.01-1.63)	0.05	1.35 (0.91-2.00)	0.14
Conduct Disorder	**1.95** (1.39-2.75)	≤0.001	**1.86** (1.36-2.55)	≤0.001	1.38 (0.88-2.16)	0.16
Non-clinical depressive episode	1.29 (0.97-1.72)	0.08	**1.40** (1.11-1.77)	0.005	**3.52** (2.13-5.83)	≤0.001
Parental alcohol problems	1.14 (0.83-1.56)	0.43	1.05 (0.81-1.38)	0.70	1.23 (0.80-1.90)	0.34
Parental Drug Problems	1.64 (0.92-2.91)	0.09	1.57 (0.91-2.69)	0.10	0.89 (0.43-1.88)	0.77
Single parent family	0.93 (0.54-1.61)	0.80	0.95 (0.60-1.50)	0.83	1.47 (0.71-3.08)	0.30
High parental conflict	0.85 (0.63-1.15)	0.30	0.96 (0.74-1.23)	0.74	1.40 (0.88-2.21)	0.15
Strict parenting	1.08 (0.81-1.43)	0.61	1.04 (0.83-1.31)	0.73	0.87 (0.58-1.30)	0.49
High levels of high school peer cannabis use	1.12 (0.71-1.77)	0.62	1.26 (0.67-2.35)	0.47	1.25 (0.67-2.35)	0.47
Childhood sexual abuse	1.52 (0.99-2.35)	0.06	0.94 (0.64-1.39)	0.77	1.46 (0.85-2.52)	0.17
Infrequent childhood religious attendance	1.23 (0.93-1.63)	0.15	1.04 (0.82-1.31)	0.74	0.93 (0.61-1.41)	0.73
Lifetime monthly alcohol use	0.70 (0.27-1.79)	0.45	0.85 (0.40-1.80)	0.68	0.63 (0.26-1.51)	0.30
Lifetime weekly tobacco use	**3.44** (2.25-5.28)	≤0.001	**1.94**(1.42-2.65)	≤0.001	**2.61**(1.27-5.34)	0.01
Lifetime other drug use	**5.99** (3.88-9.24)	≤0.001	**3.96** (2.98-5.25)	≤0.001	**3.40** (1.74-6.68)	≤0.001
Lifetime alcohol dependence	1.01 (0.76-1.35)	0.95	1.15 (0.89-1.47)	0.28	1.07 (0.69-1.65)	0.77
Lifetime tobacco dependence	**1.49** (1.06-2.09)	0.02	**1.84** (1.37-2.48)	≤0.001	1.16 (0.68-1.99)	0.58
Lifetime other drug drug dependence	**2.44** (1.64-3.64)	≤0.001	**2.64** (1.76-3.95)	≤0.001	**3.14** (1.90-5.21)	≤0.001
Early initiation cannabis use (≤16)	**1.50** (1.13-1.99)	0.005	**1.80** (1.43-2.28)	≤0.001	**1.56** (1.00-2.41)	0.05

.4 Appendix 4: SLAM Clinic Data Collection Materials

Patient information sheet, consent form, and full interview. Starts on next page.

What does the study involve?

If you agree take part the researcher will ask you to fill in two copies of a consent form before accompanying you to a private area. **The study involves an interview which will take 20—40 minutes to complete, depending on your answers.** This study is not related to your clinical care, and choosing to participate will not alter your clinical care in any way.

The interview asks about:

- Your early use of heroin and other drugs
- Your current drug use
- Problems you have experienced as a result of opiate use
- The reasons you came for treatment.

Participants will be reimbursed £10 cash when they **complete** the interview.

Can I take part?

Anyone can take part if they are:

- Over 18
- In treatment for opiate dependence
- On a methadone/buprenorphine prescription

You **CANNOT** take part if you are under 18 years old.

What are the possible benefits of taking part in the study?

The results of this study may help to improve identification of individuals who would benefit from early drug use interventions. There are no direct benefits to you for taking part, but you may appreciate the opportunity to reflect and gain insight into your earlier drug use, and the circumstances surrounding treatment seeking.

What are the possible disadvantages and risks of taking part?

There are no risks associated with this study. However, please be aware that the interview will involve talking about early drug use experiences, current drug use and problems encountered as a result of use.

How will my information be kept confidential?

- Your name is not written on your responses, and all information is kept private by using anonymous ID numbers. No identifying information such as addresses or the names of other people will be recorded.
- Only researchers will have access to the information in the study.
- Responses are confidential, and will not be passed on to key workers or clinic staff UNLESS you indicate that you

are going to harm yourself, harm someone else, or have caused a death in the past. Only in this situation will the researcher pass information to the clinical team.

- Paper copies of the interview responses and consent forms will be stored securely at the Institute of Psychiatry, Psychology and Neuroscience for up to 3 years before being safely destroyed.
- Computer data are password-encrypted.

What if I don't want to take part?

If you do not want to take part after reading the information sheet, please let the researcher know. If you are happy to tell us the reason why you don't wish to take part please tell the researcher, however you are under no obligation to do this. This will have no impact on your clinical care.

What will happen if I don't want to carry on with the study?

If you agree to take part, but change your mind once the interview has started, you are free to leave the study at any time, and this will have no impact on your clinical care. Please let the researcher know if you'd like to end the interview.

What will happen if I become upset during the study?

Anyone who becomes upset will be encouraged to discuss their feelings with their keyworker. The researcher will not inform clinic staff that you have become upset during the interview unless you ask them to do so.

However, the researcher will have to inform staff if you indicate that you are going to harm yourself, harm someone else, or have caused a death in the past.

Anyone taking part is free to end the interview at any time.

What will happen to the results of this study?

Results will be analysed as part of a doctoral project, will be published in academic journals and may be presented at conferences and talks. Participants will not be individually identifiable, as all responses will be presented as a group.

Who is organising and funding this study?

The study is funded through a PhD Studentship paid for by the Medical Research Council and the Institute of Psychiatry, Psychology and Neuroscience (IoPPN). The study is organised by PhD Student Lindsey Hines and Professor Michael Lynskey, both at the IoPPN Addictions Department.

Who has reviewed this study?

The study was reviewed by the IoPPN Research and Development Department, and by the NHS Research Ethics Committee.

How have patients and the public been involved in this study?

Drug use service users and ex-service users at the Aurora Project (Lambeth) were consulted about the content of the interview.

Further information and contact details

For more information or if you have any questions please email Lindsey Hines at lindsey.hines@kcl.ac.uk.

Complaints

If you have any complaints about the study or the behaviour of the researcher please contact the study supervisor michael.lynskey@kcl.ac.uk.

Centre Number: 0

Study Number: 15/LO/0705

Participant Identification Number for this trial: 0__ __ __

CONSENT FORM

Title of Project: Early Drug Use and Later Dependence Severity in Opiate Users

Name of Researcher: Lindsey Hines

Please initial box

1. I confirm that I have read the information sheet dated 4 June 2015 (version 3) for the above study. I have had the opportunity to consider the information, ask questions and have had these answered satisfactorily. ☐

2. I understand that my participation is voluntary and that I am free to withdraw at any time without giving any reason, without my medical care or legal rights being affected. ☐

3. I understand that the information collected about me will be used to support other research in the future, and may be shared anonymously with other researchers. ☐

4. I understand that relevant sections of my medical notes and data collected during the study, may be looked at by individuals from King's College London, from regulatory authorities or from the NHS Trust, where it is relevant to my taking part in this research. I give permission for these individuals to have access to my records. ☐

5. I agree to take part in the above study. ☐

Name of Participant	Date	Signature
Lindsey Hines		
Name of person taking consent	Date	Signature

Anonymous ID: 0 __ __ __

An Exploratory Study of Early Drug Use and Later Dependence Severity in Opiate Users

- This interview will focus on your early drug use, your treatment seeking, and dependence severity. The questions are mostly closed (e.g. yes/no) which mean you should be able to give a one or two word answer to each one.
- Responses are confidential, and will not be passed on to key workers or clinic staff UNLESS you indicate that you may harm yourself, harm someone else, or have caused a death in the past. Only in this situation will I pass information to the clinical team.
- If you agree to take part, but change your mind once the interview has started, you are free to end the interview at any time, and this will have no impact on your clinical care. Please let me know if you'd like to end the interview.
- If you become upset, I'll encourage you to discuss this with your keyworker. I won't inform clinic staff that you have become upset during the interview unless you ask me to do so.

Anonymous ID: 0 __ __ __

Age at questionnaire: ___ ___	Date of questionnaire: ___ ___/___ ___/___ ___ ___ ___
Sex	Male
	Female
Ethnicity	White British
	White Irish
	Other White
	White & Black Caribbean
	White & Black African
	White & Asian
	Other Mixed
	Indian
	Pakistani
	Bangladeshi
	Other Asian
	Caribbean
	African
	Other Black
	Chinese
	Other
Highest level of education	High school
	Sixth Form/College
	University
Employment status	Unemployed
	Part time
	Full time
	Student
	Volunteer
Marital status	Married
	Cohabiting
	Single

Anonymous ID: 0 __ __ __

Primary drug used	Opiates Only	If so, is heroin the sole opiate? YES NO
	Opiates & Crack	If so, is heroin the sole opiate? YES NO
	Crack Only	
	Benzodiazepines	
	Amphetamines (excl. Ecstasy)	
	Cocaine (excl. Crack)	
	Hallucinogens	
	Ecstasy	
	Cannabis	
	Solvent	
	Barbiturates	
	Major Tranquilisers	
	Anti-depressants	
	Other Drugs	
	Polydrug	
	Prescription Drugs	
	Misuse free/unknown	
Current methadone/buprenorphine dosage		
Length of time in prescribing	Less than 12 months	
	1 to 2 Years	
	2 to 3 Years	
	3 to 4 Years	
	4 to 5 Years	
	5+ years	
Referred to treatment by	A&E	
	Arrest Referral/DIP	
	CARAT/Prison	
	Community Care Assessment	
	Connexions	
	DRR	
	Drug Service Non-statutory	
	Drug Service Statutory	
	Education Service	
	Employment Service	
	GP	
	Probation	
	Psychiatry	
	Self	
	Social Services	
	Syringe Exchange	

Anonymous ID: 0 __ __ __

Current treatment type	Prescribing (including key working)
	Structured Intervention
	Psychosocial
	Structured Day Programme (SDP)
	Prescribing (inc. key working) and psychosocial
	Prescribing (inc. key working) and Structured Day Programme
	Inpatient detoxification (IP)
	Residential Rehabilitation (RR)
	Prescribing (inc. key working) and IP
	Prescribing (inc. key working) and RR
	Prescribing(inc. key working), psychosocial/SDP and RR
	Psychosocial/SDP and RR
	Prescribing (inc. key working), SDP and psychosocial
	All other combinations
	No adult modality
Receiving any other mental health treatment?	Yes
	No

Anonymous ID: 0 __ __ __

Transition speeds

		Alcohol	Tobacco	Cannabis	Cocaine	Heroin
Have you ever used?						
Age at first opportunity to use: "By an opportunity I mean someone either offered you [drug], or you were present when others were using and you could have used if you wanted to. Thinking back your entire lifetime, how old were you the very first time you had an opportunity to use [drug]?" (Storr et al 2011)						
Age when first used						
How quickly following opportunity? Record whatever information participant can provide	*Days*					
	Weeks					
	Months					
	Years					
Age at second use						
How quickly following first use? Record whatever information participant can provide	*Days*					
	Weeks					
	Months					
	Years					

Anonymous ID: 0 __ __ __

Who were you with when:

	First opportunity to use heroin	First used heroin	Second use heroin
Spouse/partner, boyfriend/girlfriend			
Parent			
Sibling			
Other relative			
Friend			
Acquaintance			
Stranger			
Step or foster parent			

Looking back on your heroin use, what do you consider to be the first sign that you were having problems?

_____

_____

_____

How old were you at this time?

Anonymous ID: 0 __ __ __

	Age
First solitary use	
First daily use	
First experiencing withdrawal symptoms	
First experiencing relationship problems as a result of heroin use	
First experiencing problems with employment/studies as a result of heroin use	
First injecting (if ever)	
Period of heaviest use	

Have you used other opiates aside from heroin, e.g. illegal use of prescription medications.

YES / NO

At your period of heaviest use, what amount of heroin/opiates were you using on a typical day?

Heroin:

Other opiates:

At your period of heaviest use, how many days each month were you using opiates?

Heroin:

Other opiates:

Anonymous ID: 0 __ __ __

How much do you agree with the following statements relating to the **PRESENT – referring to opiates other than methadone**:

Do you think your use of opiates is out of control?				
	0	1	2	3
	Never/Almost never	Sometimes	Often	Nearly always/Always
Does the prospect of missing a fix (or dose) or not chasing make you anxious or worried?				
	0	1	2	3
	Never/Almost never	Sometimes	Often	Nearly always/Always
D0 you worry about your use of opiates?				
	0	1	2	3
	Never/Almost never	Sometimes	Often	Nearly always/Always
Do you wish you could stop using opiates?				
	0	1	2	3
	Never/Almost never	Sometimes	Often	Nearly always/Always
How difficult do you find it to stop or go without opiates?				
	0	1	2	3
	Not difficult	Quite difficult	Very difficult	Impossible

Which of the following have you encountered as a result of your opiate use?

Issue	Yes	No	
Overdose (number)			Number of instances:
Femoral injecting			
Neck injecting			

Anonymous ID: 0 __ __ __

Treatment

How many OPIATE treatment episodes have you had?

At what age did you first seek help from a professional for your opiate use?

Why did you seek help at this time?

_____

_____

_____

_____

_____

_____

_____

_____

Looking back, should you have sought help earlier than you did?

YES / NO

If yes, what age should you have sought help?

At what age did you first receive treatment for your opiate use?

Anonymous ID: 0 __ __ __

Think back to the time you first sought treatment for your opiate use. How much do you agree with the following statements relating to your opiate use when you sought treatment for the **FIRST** time:

Did you think your use of opiates was out of control?				
	0	1	2	3
	Never/Almost never	Sometimes	Often	Nearly always/Always
Did the prospect of missing a fix (or dose) or not chasing made you anxious or worried?				
	0	1	2	3
	Never/Almost never	Sometimes	Often	Nearly always/Always
Did you worry about your use of opiates?				
	0	1	2	3
	Never/Almost never	Sometimes	Often	Nearly always/Always
Did you wish you could stop using opiates?				
	0	1	2	3
	Never/Almost never	Sometimes	Often	Nearly always/Always
How difficult did you find it to stop or go without opiates?				
	0	1	2	3
	Not difficult	Quite difficult	Very difficult	Impossible

.5 Appendix 5: Early Onset Sensitivity Tests

Table of MZ/DZ male and female and opposite sex twin pair correlations, testing sensitivty of the early cannabis use opportunity cut off point used in Chapter 4.

	Opportunity/Abuse and/or dependence twin 1/twin 2		Age of opportunity twin 1/twin 2		Abuse and/or dependence twin 1/twin 2	
Early Opportunity Cut Point	Correlation	95% CI	Correlation	95% CI	Correlation	95% CI
14 and under						
MZ males	0.37	(0.30 – 0.44)	0.76	(0.60 – 0.87)	0.72	(0.61 – 0.85)
DZ males	0.36	(0.11 – 0.57)	0.31	(-0.06 – 0.41)	0.44	(0.18 – 0.53)
MZ females	0.47	(0.34 – 0.55)	0.67	(0.57 – 0.69)	0.84	(0.72 – 0.92)
DZ females	0.15	(0.00 – 0.30)	0.48	(0.47 – 0.54)	0.47	(0.23 – 0.67)
DZ opposite sex	0.39	(0.26 – 0.56)	0.20	(0.02 – 0.37)	0.35	(0.09 – 0.41)
15 and under						
MZ males	0.41	(0.26 – 0.54)	0.72	(0.57 – 0.83)	0.72	(0.52 – 0.85)
DZ males	0.47	(0.46 – 0.63)	0.48	(0.22 – 0.68)	0.46	(0.16 – 0.69)
MZ females	0.42	(0.30 – 0.53)	0.63	(0.54 – 0.71)	0.83	(0.71 – 0.85)
DZ females	0.17	(0.02 – 0.31)	0.42	(0.28 – 0.54)	0.48	(0.39 – 0.63)
DZ opposite sex	0.23	(0.06 – 0.37)	0.26	(0.09 – 0.42)	0.33	(0.07 – 0.37)
16 and under						
MZ males	0.44	(0.31 – 0.56)	0.71	(0.59 – 0.80)	0.73	(0.53 – 0.86)
DZ males	0.39	(0.18 – 0.57)	0.55	(0.33 – 0.71)	0.36	(0.03 – 0.63)
MZ females	0.45	(0.35 – 0.55)	0.61	(0.53 – 0.69)	0.83	(0.72 – 0.92)
DZ females	0.18	(0.14 – 0.32)	0.50	(0.37 – 0.60)	0.46	(0.21 – 0.66)
DZ opposite sex	0.26	(0.10 – 0.40)	0.31	(0.14 – 0.46)	0.34	(0.08 – 0.57)

Table of associaiton between speed of early heroin transitions and later heroin use outcomes, testing sensitivity of cut points used in Chapter 6.

Transition	Speed of transition	Overdose OR (95% CI)	Groin/Neck Injecting OR (95% CI)
Early opportunity to use heroin	15 and under N = 18	2.53 (0.82 - 7.81)	4.36 (1.41 - 13.55)
	16 and under N = 27	3.23 (1.20 - 8.66)	3.74 (1.45 - 9.64)
	17 and under N = 31	3.29 (1.35 - 9.00)	3.55 (1.44 - 8.77)
	18 and under N = 40	2.08 (0 .89 - 4.84)	3.85 (1.62 - 9.18)
	19 and under N = 42	2.43 (1.04 - 5.67)	4.76 (1.97 -11.48)
Time from opportunity to initiation	Less than a week N = 57	1.05 (0.44 - 2.54)	0.72 (0.30 - 1.74)
	Within a week N = 58	0.94 (0.39 - 2.29)	0.66 (0.27 - 1.60)
	Within 3 months N = 64	0.86 (0.33 - 2.23)	1.09 (0.42 - 2.82)
	Within 1 year N = 64	0.86 (0.33 - 2.24)	1.09 (0.42 - 2.82)
	More than 1 year N = 71	0.63 (0.21 - 1.88)	1.01 (0.38 - 3.24)
Time from initiation to subsequent heroin use	Less than a week N = 47	2.10 (0.92 - 4.82)	2.13 (0.92 - 4.90)
	Within a week N = 58	1.81 (0.77 - 4.22)	1.58 (0.67 - 3.72)
	Within 3 months N = 72	3.14 (1.13 - 8.75)	1.38 (0.51 - 3.72)
	Within 1 year N = 74	3.36 (1.15 - 9.84)	1.56 (0.52 - 4.12)
	More than 1 year N = 78	2.88 (0.90 - 9.21)	1.22 (0.40 - 3.76)

.6 Papers Published from Thesis

1) The association between speed of transition from initiation to subsequent use of cannabis and later problematic cannabis use, abuse and dependence
2015, Addiction. 110, 8, p. 1311-13202
2) Genetic and Environmental Interplay in Adolescent Substance Use Disorders
2015, Current addiction reports. 2, 2, p. 122-129
3) Onset of opportunity to use cannabis and progression from opportunity to dependence: Are influences consistent across transitions?
2016. Drug and Alcohol Dependence. 160, p. 57-64

Addiction
SSA SOCIETY FOR THE STUDY OF ADDICTION
RESEARCH REPORT
doi:10.1111/add.12963

The association between speed of transition from initiation to subsequent use of cannabis and later problematic cannabis use, abuse and dependence

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ABSTRACT

Aims To test whether speed of transition from initiation use to subsequent use of cannabis is associated with likelihood of later cannabis dependence and other outcomes, and whether transition speed is attributable to genetic or environmental factors. **Design** Cross-sectional interview study. **Setting** Australia. **Participants** A total of 2239 twins and siblings who reported using cannabis at least twice [mean age at time of survey = 32.0, 95% confidence interval (CI) = 31.9 – 32.1, range = 22–45]. **Measurements** Time between initiation and subsequent cannabis use (within 1 week; within 3 months; between 3 and 12 months; more than 1 year later), later use of cannabis and symptoms of DSM-IV cannabis abuse/dependence. Multinomial regression analyses (comparison group: more than 1 year later) adjusted the association between speed of transition and the outcomes of cannabis daily use, abuse/dependence and treatment-seeking after controlling for socio-demographic, childhood, mental health, peer and licit drug factors. Twin modelling estimated the proportion of variance in transition speed attributable to genetic (A), common environment (C) and unique environmental (E) factors. **Findings** Subsequent use of cannabis within 1 week of initiation was associated with daily use [odds ratio (OR) = 2.64, 95% CI = 1.75–3.99], abuse and/or dependence (OR = 3.25, 95% CI = 2.31–4.56) and treatment-seeking for cannabis problems (OR = 1.89, 95% CI = 1.03–3.46). Subsequent use within 3 months was associated with abuse and/or dependence (OR = 1.61, 95% CI = 1.18–2.19). The majority of the variation of the speed of transition was accounted for by unique environment factors (0.75). **Conclusions** Rapid transition from initiation to subsequent use of cannabis is associated with increased likelihood of subsequent daily cannabis use and abuse/dependence.

Keywords Cannabis, cannabis abuse, cannabis dependence, initiation, subsequent use, transitions, twin study.

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INTRODUCTION

Cannabis is the most commonly used illicit drug, with prevalence of life-time use estimated at between 2.7 and 4.9% of the global population aged 15–64 years [1]. Although many individuals use cannabis infrequently and without problematic consequences, globally an estimated 13.1 million individuals experience cannabis dependence, contributing 10.3% of the illicit drug use global burden of disease [2].

Existing research has identified a number of genetic and environmental factors associated with increased risks for cannabis dependence [3–12]. However, a number of intermediate stages of use occur necessarily before an individual develops dependence. These include opportunity to use, initiation, repeated use and escalation to regular use, and genetic and environmental factors are associated differentially with progression through these stages [8,10,12–15].

Less is known about variation in progression through the stages of substance use. Research in this area focuses on speed of transition, including speed from initiation of use to: daily use [16]; regular use [17]; and abuse or dependence [17–19]. More research has focused on early onset of use, which can be used as an exemplar of the speed of

transition literature by representing early onset of drug use as a faster rate of transition from non-use to initiation. This is associated with alcohol, tobacco and cannabis dependence [18,20–22], suggesting a relationship between rate of transition and later substance use outcomes. Given that there is thought to be a short period after substance use initiation for implementation of prevention interventions [23,24], the potential for speed of transition to act as an early marker for later problems is a worthwhile avenue for exploration.

The relationship between transition speed and later drug-use outcomes is not straightforward. Those at risk of dependence may be expected to begin and continue on a faster trajectory through the stages of substance use, but research demonstrates that those who progress faster from non-use to initiation often exhibit a slower progression to dependence than those who experience later initiation [18,25]. Additionally, faster transition from initiation to regular use has not been associated consistently with later outcomes of dependence [17]. Further research on a broader range of transitions is required to understand more clearly the relationship between speed of transition and later outcomes, and to identify whether similar factors determine speed between each stage [13].

One previously unstudied transition is that from initiation (first use) to the subsequent (second) use of cannabis. Utilizing cross-sectional data from a sample of Australian twins, this paper aims to:

1) Test whether speed of transition from initiation to subsequent use of cannabis is associated with increased likelihood of later daily cannabis use, abuse and/or dependence and cannabis-related treatment-seeking when accounting for the influence of socio-demographic, childhood, mental health, peer and licit drug factors that may be predictive of faster transitions in the subsequent use of cannabis.
2) Determine the extent to which the speed of this transition is attributable to additive genetic, shared environmental or non-shared environmental factors.

METHODS

Sample

From Australian Twin Registry members born between 1972 and 1979, 3348 monozygotic (MZ) and dizygotic (DZ) twins and 476 of their siblings completed a drug misuse study (see [26] for a recruitment outline). Of the complete cohort sample, 2601 (68.5%) reported life-time use of cannabis. The subset of the sample selected for the analyses in this paper were the 2239 participants [mean age at time of survey = 32.0, 95% confidence interval (CI) = 31.9–32.1, range = 22–45] who had reported using cannabis at least twice in their lives (58.6% of the entire sample, 86.1% of life-time cannabis users). Of this subset, 58.7% were female.

Assessment

Participants were assessed through computer-assisted telephone interviews, and were provided with a respondent booklet so that answers would be unidentifiable to anyone overhearing. The interview collected information on socio-demographics, childhood experiences, substance use and common mental health disorders, including conduct disorder, assessed using the Semi-Structured Assessment for the Genetics of Alcoholism (SSAGA)-II interview [27]. The SSAGA is a validated measure of mental health that uses DSM-IV criteria, and includes alcohol and other drug abuse and dependence.

Measures

Transition speed

Those who reported using cannabis more than once were asked: 'How soon after you first tried marijuana did you try it again?'. Data were recorded categorically, and responses were further collapsed for analysis into the following categories: within 1 week (19.8%), within 3 months (but not including those who transitioned within 1 week) (37.7%), between 3 months and 12 months (21.7%) and more than 1 year later (20.8%).

Life-time cannabis involvement

Daily use of cannabis

In the subsample used in this analysis 16.6% self-reported using cannabis daily during their period of heaviest use.

Cannabis abuse and/or dependence

In the subsample used in this analysis, 27.9% reported cannabis abuse and/or dependence. Participants were classified as meeting DSM-IV criteria for life-time cannabis abuse if they reported one or more of the following: often using cannabis in a situation where they might get hurt; arrested more than twice within a 12-month period as a result of their cannabis use; cannabis use having caused difficulty with work, study or household responsibilities; and cannabis having caused social and interpersonal problems more than three times within a 12-month period.

Participants were classified as meeting life-time criteria for DSM-IV cannabis dependence if they reported three or more of the following symptoms occurring within the same 12-month period: using cannabis a greater number of times/greater amount than was intended, tolerance, wanting to cut down/stop use, spending so much time obtaining/using/recovering from the effects of cannabis

that the participant had little time for anything else, reducing important activities as a result of cannabis use and continuing use despite it worsening health/emotional problems. Withdrawal was not included, as it was not part of DSM-IV criteria for cannabis dependence.

Cannabis-related treatment-seeking

In the subsample used in this analysis, 6% self-reported having discussed cannabis-related problems with a professional. Participants were able to endorse seeking treatment from multiple sources: psychiatrist ($n = 45$), general practitioner or other medical doctor ($n = 80$), psychologist ($n = 42$), another mental health professional ($n = 61$), member of the clergy ($n = 7$) or another source ($n = 9$).

Covariates

Early cannabis onset

Individuals reporting life-time cannabis use were asked the age at which they first used cannabis. In line with existing literature [26,28,29], those who were aged 16 and under when cannabis was first used were classified as having early onset of cannabis use. Additionally, a series of sensitivity tests were conducted to test the effect of different early-onset cut-off points (< 13, < 14, < 15 and < 17), which showed that selecting 16 as the cut-off had no effect on the results of the analyses (full results available upon request). Mean age of cannabis onset in the analytical sample was 17.46 [standard deviation (SD) = 2.99] with a range of 6–34 years.

Education

Participants were asked to report the highest level of education they had obtained, and for analysis respondents were classified by whether or not their highest level of education was post-secondary/higher education.

Parental characteristics

Parental alcohol problems were determined through participant self-report of their mother or father's problems with health/family/job/police/other as a result of drinking, or their mother or father drinking excessively. Specifically, participants were asked: 'Did drinking ever cause [your biological father/mother] to have problems with health, family, job or police, or other problems?' and 'Did you ever feel that [your biological father/mother] were excessive drinkers?'. Responding 'yes' to either of these questions constituted being a case for parental alcohol problems.

Parental drug problems were determined through participant self-report of their mother or father's problems with health/family/job/police/other as a result of drug use, or the participant reporting that they felt their mother or father had a problem with drugs. Specifically, participants were asked: 'Did using drugs ever cause [your biological father/mother] to have problems with health, family, job or police, or other problems?' and 'Did you ever feel that [your biological father/mother] had a problem with drugs?'. Responding 'yes' to either of these questions constituted being a case for parental drug problems.

Parental conflict was determined by participant responses to the questions: 'How often did your parents fight or argue in front of you?' and 'How much conflict and tension was there between your parents?'. Both questions focused on the period when the participant was aged 6–13 years. Participants reporting parents 'sometimes' or 'always' fought or argued, or reporting 'a lot' or 'some' conflict/tension, were coded as experiencing high parental conflict.

Childhood sexual abuse

Participants who self-reported being forced into sexual intercourse or any other forms of sexual activity before age 18 were classified as having experienced childhood sexual abuse.

Conduct disorder

Participants were coded as meeting criteria for conduct disorder if they reported at least three of the 15 DSM-IV criteria occurring within the same 12-month period, prior to age 18.

Depressed mood before cannabis onset

Participants were classified as having experienced depressed mood if they had reported feeling depressed/down/low 'most of the day' and 'nearly every day', or feeling a great deal less interested in or able to enjoy most things 'most of the day' and 'nearly every day' for at least 2 weeks in their life-time before the onset of cannabis use.

Peer use

The extent of substance misuse among high school peers was measured through self-report questions asking whether 'hardly any', 'some', 'half', 'three-quarters' or 'almost all' the students who were in their grade in high-school used illegal drugs while of school age. Participants were categorized as being exposed to high levels of illicit drug use during high school if they reported that at least three-quarters of their peers had been using cannabis.

Regular alcohol use before cannabis onset

Age of onset of regular alcohol use (once a month for 6 months or longer) and age of cannabis onset were used to determine whether regular alcohol use occurred before onset of cannabis use.

Regular tobacco use before cannabis onset

The age of onset of regular tobacco use (at least once a week for at least 2 months) and age of cannabis onset were used to determine whether regular tobacco use occurred before onset of cannabis use.

Statistical analysis

Epidemiological analyses were conducted in SAS statistical software version 9.3 for Windows (SAS Institute Inc., Cary, NC, USA) and Stata statistical software version 11 (StataCorp, College Station TX, USA, 2009). χ^2 tests and phi coefficients assessed the association between the speed of transition from initiation to subsequent use of cannabis and life-time cannabis daily use, abuse and/or dependence and treatment-seeking. All associations were deemed significant at the $P < 0.05$ level. Multinomial logistic regression analysis (reference category: subsequent use more than a year after initiation) determined the association between the speed of transition from initiation to subsequent use of cannabis and the outcomes daily cannabis use, abuse/dependence and treatment-seeking for cannabis use problems after adjustment for socio-demographic, childhood, mental health, peer and licit drug factors. Covariates were included in the models if they were associated significantly with both the exposure and outcome variables through χ^2 tests (analyses not reported). To correct for the non-independence of observations, Huber–White analysis for clustered data was implemented in Stata to provide robust standard errors. *Post-hoc* comparisons across the varying speeds of transition were conducted using Wald χ^2 tests.

Twin modelling was conducted using OpenMX [30] for the statistical software R [31]. As there were low numbers of concordant twins, univariate analyses used raw ordinal data and full-information maximum-likelihood (FIML) estimation, which makes use of twin pairs where data from a co-twin is unavailable. Composition of the twin sample is described in Table 1. Model-fitting was conducted using a stepwise approach. A liability-threshold model including an adjustment for twin sex and estimating co-twin correlations was fitted to the data set and used to test assumptions regarding the equality of thresholds within and between MZ and DZ twin groups. Based on these results, a univariate variance components model was fitted, partitioning the variance attributable to additive genetic (A), shared environmental (C) and unique environmental (E) factors. Difference in model fit was assessed via the likelihood-ratio χ^2 test and examination of the Akaike information criterion (AIC) and Bayesian information criterion (BIC).

RESULTS

Associations between speed of transition and daily use, abuse/dependence and treatment-seeking

Speed of transition was associated significantly with each of the three cannabis use outcomes ($P < 0.0001$ for all outcomes; see Table 2). Those whose second use of cannabis was within 1 week of initiation had the highest rate of daily cannabis use (28.4%), abuse and/or dependence (46.0%) or cannabis-related treatment-seeking (10.6%). For all outcomes, the proportion that would go on to develop problems decreased approximately linearly across the groups.

Demographic, childhood and peer use associations with transition speed

Significant differences were observed between the different transition speed groups for almost all the socio-demographic, childhood, mental health, peer and licit drug factors tested in this analysis (see Table 3). Parental drug problems, parental conflict and depressed mood before cannabis onset were not associated significantly with transition speed.

Multinomial logistic regression of the outcomes associated with transition speed

After controlling for early onset of cannabis use, socio-demographic, childhood, mental health, peer and licit

Table 1 Speed of transition from initiation to subsequent use of cannabis by zygosity for twin analysis sample.

Twin sample	*Within a week n = 400*	*Within 3 months n = 746*	*3 months to 1 year n = 412*	*More than a year n = 411*
MZ twin 1	73	145	83	94
n = 395	18.5%	36.7%	21.0%	23.8%
MZ twin 2	99	147	90	93
n = 429	23.1%	34.3%	20.9%	21.7%
DZ twin 1	101	235	126	113
n = 575	17.6%	40.9%	21.9%	19.6%
DZ twin 2	127	219	113	111
n = 570	22.3%	38.4%	19.8%	19.5%

DZ = dizygotic; MZ = monozygotic.

Table 2 Association between speed of transition from initiation to subsequent cannabis use and cannabis-related outcomes.

Variable	*More than a year n = 465*	*3 months to 1 year n = 487*	*Within 3 months n = 844*	*Within a week n = 443*	*Phi*	*P-value*
Daily use	45	67	134	126	0.17	<0.0001
n = 372	9.7%	13.8%	15.9%	28.4%		
Abuse and/or dependence	82	100	238	204	0.22	<0.0001
n = 624	17.6%	20.5%	28.2%	46.0%		
Treatment-seeking	19	21				<0.0001
n = 132	45	47	0.10	10.6%		

Table 3 Associations between speed of transition from initiation to subsequent cannabis use and socio-demographic, childhood, mental health, peer and licit drug factors.

Variable	*More than a year n = 465*	*3 months to 1 year n = 487*	*Within 3 months n = 844*	*Within a week n = 443*	*Phi*	*P-value*
Mean age at cannabis initiation	17.60 (SD = 2.95)	17.94 (SD = 3.16)	17.24 (SD = 2.89)	17.23 (SD = 2.98)	0.20	0.1009
Gender: female	297	289	489	239	0.06	0.0230
n =1314	63.9%	59.3%	57.9%	53.9%		
Education: any high school	98	131	207	159	0.11	<0.0001
n = 595	21.1%	26.9%	24.5%	35.9%		
Parental alcohol problems	137	118	225	147	0.07	0.0082
n = 627	30.0%	24.6%	27.1%	34.3%		
Parental drug problems	19	21	36	30	0.05	0.1528
n = 106	4.2%	4.4%	4.3%	6.9%		
Parental conflict	202	176	321	185	0.05	0.0925
n = 884	45.3%	38.1%	41.0%	44.6%		
Experienced sexual abuse before age 18	53	40	80	59	0.06	0.0504
n = 232	11.4%	8.2%	9.5%	13.4%		
Conduct disorder	35	46	106	98	0.15	<0.0001
n = 285	7.5%	9.4%	12.6%	22.1%		
Depressed mood before cannabis onset	29	51	73	46	0.05	0.0778
n = 199	6.2%	10.5%	8.7%	10.4%		
Peer use: more than ¾ of high school peers used cannabis	38	28	87	56	0.08	0.0020
n = 209	8.2%	5.7%	10.3%	12.6%		
Early cannabis onset: 16 and under	178	173	380	198	0.08	0.0016
n = 929	38.3%	35.5%	45.0%	44.7%		
Regular nicotine use before cannabis onset	80	96	151	123	0.10	<0.0001
n = 450	17.2%	19.7%	17.9%	27.8%		
Regular alcohol use before cannabis onset	165	182	256	127	0.07	0.0077
n = 730	35.5%	37.4%	30.3%	28.7%		

drug factors, those whose second use of cannabis was within a week were at increased odds of meeting criteria for abuse/dependence [odds ratio (OR) = 3.25, 95% confidence interval (CI) = 2.31–4.56], reporting daily use (OR = 2.64, 95% CI = 1.75–3.99) and treatment-seeking (OR = 1.89, 95% CI = 1.03–3.46) (see Table 4). Those whose subsequent use of cannabis was within 3 months of initiation were just under twice as likely to develop abuse and/or dependence (OR = 1.61, 95% CI = 1.18–2.19).

Post-hoc analysis of age of onset

Stratifying the analysis by early or later onset revealed differences in the association between transition speed and all later outcomes, which remained after adjustment

Table 4 Odds ratios (95% confidence intervals) between speed of transition from initiation to subsequent cannabis use, covariates and later cannabis outcomes from multinomial logistic regression.

		Daily use[a] *n = 372 odds ratio (95% confidence interval)*		*Abuse and/or dependence n = 624 odds ratio (95% confidence interval)*		*Treatment-seeking*[a] *n = 132 odds ratio (95% confidence interval)*	
Outcome		*Univariate model*	*Adjusted model*	*Univariate model*	*Adjusted model*	*Univariate model*	*Adjusted model*
Speed of transition to subsequent use	More than a year, *n* = 465	1.00	1.00	1.00	1.00	1.00	1.00
	3 months to 1 year, *n* = 487	1.49* (1.00–2.21)	1.43 (0.95–2.17)	1.21 (0.87–1.67)	1.19 (0.85–1.68)	1.06 (0.58–1.94)	1.03 (0.55–1.90)
	Within 3 months, *n* = 844	1.76* (1.23–2.52)	1.44 (0.99–2.11)	1.83*** (1.37–2.46)	1.61** (1.18–2.19)	1.32 (0.76–2.28)	1.05 (0.60–1.84)
	Within a week, *n* = 443	3.71*** (2.55–5.39)	2.64*** (1.75–3.99)	3.99*** (2.92–5.44)	3.25*** (2.31–4.56)	2.79*** (1.60–4.85)	1.89* (1.03–3.46)
Covariates							
Gender: female *n* = 1314		0.54*** (0.41–0.71)		0.47*** (0.37–0.59)		0.60** (0.40–0.92)	
Education: any high school *n* = 595		1.47* (1.12–1.93)		1.23 (0.97–1.55)		1.41 (0.98–2.03)	
Parental alcohol problems *n* = 627		1.20 (0.90–1.60)		1.15 (0.90–1.47)		1.46 (0.97–2.17)	
Conduct disorder *n* = 285		2.67*** (1.95–3.65)		2.55*** (1.89–3.45)		2.50*** (1.61–3.90)	
Peer use: more than ¾ of high school peers used cannabis *n* = 209		1.14 (0.76–1.70)		0.93 (0.65–1.31)		1.42 (0.82–2.46)	
Early cannabis onset: 16 and under *n* = 929		2.00*** (1.50–2.66)		2.21*** (1.72–2.84)		1.68* (1.05–2.70)	
Regular nicotine use before cannabis onset *n* = 450		1.59** (1.16–2.17)		1.44** (1.10–1.87)		Not included in model	
Regular alcohol use before cannabis onset *n* = 730		0.51*** (0.36–0.74)		0.60*** (0.45–0.80)		0.60 (0.34–1.09)	
Experienced sexual abuse before age 18 *n* = 232		1.99*** (1.34–2.95)		2.00*** (1.41–2.85)		2.03** (1.18–3.48)	

*$P < 0.05$; **$P < 0.01$; ***$P < 0.001$. [a]For these outcomes the groups '3 months to 1 year' and 'within 3 months' were not found to be significantly different to each other in *post-hoc* tests.

for the other covariates. For the association between transitions within a week and daily use, those with earlier onset had an increase in likelihood of 1.83 (95% CI = 1.05–3.17) compared to 4.32 (95% CI = 2.27–8.21) for those with later onset.

For the association between transitions within a week and abuse/dependence, those with earlier onset had an increase in likelihood of 2.14 (95% CI = 1.33–3.42) compared to 4.86 (95% CI = 2.97–7.94) for those with later onset. For the association between transitions within a week and later treatment-seeking, those with earlier onset had an increase in likelihood of 1.63 (95% CI = 0.72–3.70) compared to 2.19 (95% CI = 0.92–5.17) for those with later onset.

There was a significant interaction between early/later cannabis onset and (1) transition within a week, with those in the early-onset group having a decrease in likelihood of abuse and/or dependence of 0.50 (95% CI = 0.26–0.94) and (2) transition 3 months–1 year, with those in the early-onset group having an increase in likelihood of daily use (OR = 2.55, 915% CI = 1.04–6.27) and treatment-seeking (OR = 8.38, 95% CI = 1.35–2.1).

Modelling additive genetic, shared and non-shared environmental influences on speed of transition between initiation and subsequent cannabis use

Data on speed of transition from initiation to subsequent use of cannabis for twin modelling was available for 824 MZ twins and 1145 DZ twins (see Table 1 for full information). Tetrachoric correlations were similar for MZ (0.27) and DZ (0.23) pairs. A univariate variance component twin model was fitted, with thresholds equated within and between zygosity groups, as initial analyses did not identify any significant differences ($P = 0.17$). The estimate for additive genetic influences for the full model was small (0.002, 95% CI = 1.446372e-09–0.35), and could be dropped from the model without a significant loss of fit ($P = 1$). A model specifying only environmental influences (C and E) provided the best fit, with moderate shared environmental influences (0.25, 95% CI = 0.15–0.34) and large unique environmental influences (0.75, 95% CI = 0.66–0.84) on the variation in speed of this transition (see Table 5).

DISCUSSION

The key finding of this paper was the significant association between speed of transition from initiation to subsequent use of cannabis and later likelihood of daily cannabis use, cannabis abuse/dependence and cannabis-related treatment-seeking. This association remained after controlling for potential confounders. The unique environment accounted for most (0.75) of the variance in the speed of transition from initiation of cannabis to subsequent use, and measured risk factors including conduct disorder, education and regular use of nicotine before cannabis initiation were associated with a more rapid transition to subsequent use. Given the absence of prior research on this transition, these findings provide an original and intriguing contribution to the literature.

Previous research has found that earlier use is associated with later problematic drug use/dependence [18,21,22,32–34], and by studying the novel transition from initiation to subsequent use this paper has established that the association between speed of transition and later negative outcomes remains after controlling for factors that would be expected to predispose individuals towards cannabis use problems. Stratifying analyses by onset showed the association between transition speed and all studied outcomes was stronger among those with later cannabis onset, suggesting that transition speed is indicative of later problems even beyond the high-risk period of early adolescence. This highlights the importance of accounting for age when applying a stage-sequential approach to the study of substance use [13].

Additive genetic effects have no influence on variation in the speed of this transition, which is in contrast to findings of moderate heritability for other transitions [5,26,28,35]. Similarly, the speed of other specific transitions has been found to be moderately heritable, with 0.30 (95% CI = 0.15–0.46) of the rate of transition from non-use to initiation attributed to additive genetic effects

Table 5 Twice ACE model fitting results and variance components point estimates with 95% confidence intervals for speed of transition from initiation to subsequent use of cannabis.

Model	*Proportion of variance*			*–2 log likelihood*	*df*	*AIC*	*BIC*
	A	*C*	*E*				
Full ACE model	0.0002 (5.801395e-08–0.35)	0.25 (2.7711648e-10–0.34)	0.75 (0.63–0.84)	5268.96	1963	1342.96	–9004.02
CE submodel	–	0.25 (0.15–0.34)	0.75 (0.66–0.85)	5268.96	1964	1340.96	–9011.30

AIC = Akaike information criterion; BIC = Bayesian information criterion. Model is adjusted for sex. A = additive genetic factors; C = common environmental factors; E = specific environmental factors.

and similar findings observed for the rate of transition from initiation to first dependence symptom (0.36, 95% CI = 0.19–0.44) and first dependence symptom to the development of dependence (0.37, 95% CI = 0.00–0.58) [36]. In contrast, our findings show the speed of transition from initiation to subsequent use of cannabis is influenced predominately by environmental factors, demonstrating the importance of utilizing a stage-sequential approach in order to understand fully how genetic and environmental factors vary throughout substance use.

Significant differences were observed between transition speed groups for measured environmental risk factors. Studies of the speed of other transitions have identified similar environmental risk factors, including childhood sexual abuse [37,38], parental substance abuse [37], peer use of substances [39,40], parental substance dependence [41] and conduct disorder [41–44]. The majority of the variance in the speed of the transition from initiation to subsequent use was attributable to the unique environment, which can represent measurement error in the analysis. However, we speculate that availability, which has been found previously to account for variation in drug use progression [45], is likely to form part of the environmental factors at play in the speed of transition from initiation to subsequent use. Further exploration is needed to understand the determinants of speed of transition from initiation to subsequent use.

Limitations and future research

First, these data were based on retrospective self-report which introduces the possibility of recall bias. Secondly, the measure of transition speed was comprised of relatively wide categories. Thirdly, there was a low number of twin pairs concordant for speed of transition from initiation to subsequent use, which was overcome through the use of raw data for the twin modelling. Ordinal analysis can result in lower power, and may result in an underestimate of the true liability correlation [46]. Fourthly, the study lacked temporal information on a number of covariates within the analysis, and including these variables in the analysis represents a cautious approach to adjustment for confounding variables which may lead to underestimation of the effect of this transition. Fifthly, while probably representative of base population [47], the prevalence of life-time cannabis use in this sample is relatively high at 68.2%, which may limit generalizability.

It is unknown whether these findings will translate to alcohol and nicotine use or to other illicit drugs, given that differences have been observed previously in the rate of transition to cannabis disorder compared to nicotine or alcohol dependence [18], but the results of the current study suggest that study of this transition across drug classes is warranted.

Implications

We suggest that faster transition from initiation to subsequent use is unlikely to have a traditional causal relationship with cannabis dependence. The association probably reflects a combination of individual and contextual factors, such as availability, that surround the rapid escalation. If replicated in prospective research, these findings may have practical utility for clinical practice, with the prospect of translation into a clinically useful marker with which to identify individuals likely to benefit from intervention. These findings have also highlighted the utility of studying different transitions in substance use to disentangle the complex aetiology of drug use disorders [13].

CONCLUSIONS

Those whose subsequent use is within 1 week have the greatest likelihood of future cannabis use problems. The novel demonstration that the speed of transition from initiation to subsequent cannabis use is predictive of later cannabis outcomes is striking, and is of potentially major importance to understanding of the development of cannabis dependence and problems. Given that the variance in the speed of this transition is due predominately to unique environmental factors, it may be that speed of the transition from initiation to subsequent use acts as a proxy measure of a number of the contextual factors that contribute to the development of addiction.

Declaration of interests

A.A. has previously received peer-reviewed funding from ABMRF/Foundation for Alcohol Research, which receives partial support from the brewing industry. J.S. is a researcher and clinician and has worked with a range of types of treatment and rehabilitation service-providers. He has also worked with pharmaceutical companies to seek to identify new or improved treatments, and also with a range of governmental and non-governmental organizations. His employer (King's College London) is registering intellectual property on an innovative medication development with which J.S. is involved, and J.S. has been named in a patent registration by a Pharma company as inventor of a potential novel overdose resuscitation product. A fuller account of J.S.'s interests is on his personal web-page of the Addictions Department at http://www.kcl.ac.uk/ioppn/depts/addictions/people/hod.aspx. J.S. is also supported by the National Institute for Health Research (NIHR) Biomedical Research Centre for Mental Health at South London and Maudsley NHS Foundation Trust and King's College London. There are no other declarations of interest from the authors of this paper.

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References

1. United Nations Office on Drugs and Crime (UNODC). World Drug Report 2014. Geneva, Switzerland: UNODC; 2014.
2. Degenhardt L., Whiteford H., Hall W. D. The Global Burden of Disease projects: what have we learned about illicit drug use and dependence and their contribution to the global burden of disease? *Drug Alcohol Rev* 2014; **33**: 4–12.
3. Fowler T., Lifford K., Shelton K., Rice F., Thapar A., Neale M. C. *et al.* Exploring the relationship between genetic and environmental influences on initiation and progression of substance use. *Addiction* 2007; **102**: 413–22.
4. Kendler K. S., Karkowski L. M., Neale M. C., Prescott C. A. Illicit psychoactive substance use, heavy use, abuse, and dependence in a US population-based sample of male twins. *Arch Gen Psychiatry* 2000; **57**: 261–9.
5. Verweij K. J. H., Zietsch B. P., Lynskey M. T., Medland S. E., Neale M. C., Martin N. G. *et al.* Genetic and environmental influences on cannabis use initiation and problematic use: a meta-analysis of twin studies. *Addiction* 2010; **105**: 417–30.
6. Clark D. B., Cornelius J. Childhood psychopathology and adolescent cigarette smoking: a prospective survival analysis in children at high risk for substance use disorders. *Addict Behav* 2004; **29**: 837–41.
7. Korhonen T., Huizink A. C., Dick D. M., Pulkkinen L., Rose R. J., Kaprio J. Role of individual, peer and family factors in the use of cannabis and other illicit drugs: a longitudinal analysis among Finnish adolescent twins. *Drug Alcohol Depend* 2008; **97**: 33–43.
8. Butters J. E. Family stressors and adolescent cannabis use: a pathway to problem use. *J Adolesc* 2002; **25**: 645–54.
9. Nelson E. C., Heath A. C., Lynskey M. T., Bucholz K. K., Madden P. A., Statham D. J. *et al.* Childhood sexual abuse and risks for licit and illicit drug-related outcomes: a twin study. *Psychol Med* 2006; **36**: 1473–83.
10. Hicks B. M., Durbin C. E., Blonigen D. M., Iacono W. G., McGue M. Relationship between personality change and the onset and course of alcohol dependence in young adulthood. *Addiction* 2012; **107**: 540–8.
11. Swendsen J., Conway K. P., Degenhardt L., Glantz M., Jin R., Merikangas K. R. *et al.* Mental disorders as risk factors for substance use, abuse and dependence: results from the 10-year follow-up of the National Comorbidity Survey. *Addiction* 2010; **105**: 1117–28.
12. Andrews J. A., Tildesley E., Hops H., Li F. The influence of peers on young adult substance use. *Health Psychol* 2002; **21**: 349–57.
13. Hines L. A., Morley K. I., Mackie C., Lynskey M. T. Genetic and environmental interplay in adolescent substance use disorders. *Curr Addict Rep* 2015; **2**: 122–129.
14. Hingson R. W., Heeren T., Winter M. R. Age of alcohol-dependence onset: associations with severity of dependence and seeking treatment. *Pediatrics* 2006; **118**: e755–63.
15. Agrawal A., Lynskey M. T. Are there genetic influences on addiction: evidence from family, adoption and twin studies. *Addiction* 2008; **103**: 1069–81.
16. Best D., Day E., Cantillano V., Gaston R. L., Nambamali A., Sweeting R. *et al.* Mapping heroin careers: utilising a standardised history-taking method to assess the speed of escalation of heroin using careers in a treatment-seeking cohort. *Drug Alcohol Rev* 2008 Mar 1; **27**: 165–70.
17. Ridenour T. A., Lanza S. T., Donny E. C., Clark D. B. Different lengths of times for progressions in adolescent substance involvement. *Addict Behav* 2006; **31**: 962–83.
18. Behrendt S., Wittchen H. U., Hofler M., Lieb R., Beesdo K. Transitions from first substance use to substance use disorders in adolescence: is early onset associated with a rapid escalation? *Drug Alcohol Depend* 2009; **99**: 68–78.
19. Behrendt S., Wittchen H. -U., Höfler M., Lieb R., Low N. C. P., Rehm J. *et al.* Risk and speed of transitions to first alcohol dependence symptoms in adolescents: a 10-year longitudinal community study in Germany. *Addiction* 2008; **103**: 1638–47.
20. Hingson R. W., Zha W. Age of drinking onset, alcohol use disorders, frequent heavy drinking, and unintentionally injuring oneself and others after drinking. *Pediatrics* 2009; **123**: 1477–84.
21. Behrendt S., Beesdo-Baum K., Hofler M., Perkonigg A., Buhringer G., Lieb R. *et al.* The relevance of age at first alcohol and nicotine use for initiation of cannabis use and progression to cannabis use disorders. *Drug Alcohol Depend* 2012; **123**: 48–56.
22. Fergusson D. M., Horwood L. J. Early onset cannabis use and psychosocial adjustment in young adults. *Addiction* 1997; **92**: 279–96.
23. Gervais A., O'Loughlin J., Meshefedjian G., Bancej C., Tremblay M. Milestones in the natural course of onset of cigarette use among adolescents. *Can Med Assoc J* 2006; **175**: 255–61.
24. Wittchen H. U., Behrendt S., Hofler M., Perkonigg A., Lieb R., Buhringer G. *et al.* What are the high risk periods for incident substance use and transitions to abuse and dependence? Implications for early intervention and prevention. *Int J Methods Psychiatr Res* 2008; **17**: S16–29.
25. DeWit D. J., Adlaf E. M., Offord D. R., Ogborne A. C. Age at first alcohol use: a risk factor for the development of alcohol disorders. *Am J Psychiatry* 2000; **157**: 745–50.
26. Lynskey M. T., Agrawal A., Henders A., Nelson E. C., Madden P. A. F., Martin N. G. An Australian twin study of cannabis and other illicit drug use and misuse, and other psychopathology. *Twin Res Hum Genet* 2012; **15**: 631–41.
27. Bucholz K. K., Cadoret R., Cloninger C. R., Dinwiddie S. H., Hesselbrock V. M., Nurnberger J. *et al.* A new, semi-structured psychiatric interview for use in genetic linkage studies: a report on the reliability of the SSAGA. *J Stud Alcohol Drugs* 1994; **55**: 149.
28. Sartor C. E., Agrawal A., Lynskey M. T., Bucholz K. K., Madden P. A., Heath A. C. Common genetic influences on the timing of first use for alcohol, cigarettes, and cannabis in young African-American women. *Drug Alcohol Depend* 2009; **102**: 49–55.

29. Agrawal A., Grant J. D., Waldron M., Duncan A. E., Scherrer J. F., Lynskey M. T. *et al.* Risk for initiation of substance use as a function of age of onset of cigarette, alcohol and cannabis use: findings in a Midwestern female twin cohort. *Prev Med* 2006; **43**: 125–8.
30. Boker S. M., Neale M. C., Maes H. H., Wilde M. J., Spiegel M., Brick T. R. *et al.* OpenMx: an open source extended structural equation modeling framework. *Psychometrika* 2011; **76**: 306–17.
31. R Core Team. *R: A Language and Environment for Statistical Computing* (internet). Vienna, Austria: *R Foundation for Statistical Computing*, 2013. Available at: http://www.R-project.org/
32. Grant J. D., Lynskey M. T., Scherrer J. F., Agrawal A., Heath A. C., Bucholz K. K. A cotwin-control analysis of drug use and abuse/dependence risk associated with early-onset cannabis use. *Addict Behav* 2010; **35**: 35–41.
33. Lynskey M. T., Heath A. C., Bucholz K. K., Slutske W. S., Madden P. A., Nelson E. C. *et al.* Escalation of drug use in early-onset cannabis users vs co-twin controls. *JAMA* 2003; **289**: 427–33.
34. Swift W., Coffey C., Carlin J. B., Degenhardt L., Patton G. C. Adolescent cannabis users at 24 years: trajectories to regular weekly use and dependence in young adulthood. *Addiction* 2008; **103**: 1361–70.
35. Kendler K. S., Neale M. C., Sullivan P., Corey L. A., Gardner C. O., Prescott C. A. A population-based twin study in women of smoking initiation and nicotine dependence. *Psychol Med* 1999; **29**: 299–308.
36. Sartor C. E., Agrawal A., Lynskey M. T., Bucholz K. K., Heath A. C. Genetic and environmental influences on the rate of progression to alcohol dependence in young women. *Alcohol Clin Exp Res* 2008; **32**: 632–8.
37. Rothman E. F., Edwards E. M., Heeren T., Hingson R. W. Adverse childhood experiences predict earlier age of drinking onset: results from a representative US sample of current or former drinkers. *Pediatrics* 2008; **122**: e298–304.
38. Sartor C. E., Waldron M., Duncan A. E., Grant J. D., McCutcheon V. V., Nelson E. C. *et al.* Childhood sexual abuse and early substance use in adolescent girls: the role of familial influences. *Addiction* 2013; **108**: 993–1000.
39. Coffey C., Lynskey M., Wolfe R., Patton G. C. Initiation and progression of cannabis use in a population-based Australian adolescent longitudinal study. *Addiction* 2000; **95**: 1679–90.
40. Blitstein J. L., Robinson L. A., Murray D. M., Klesges R. C., Zbikowski S. M. Rapid progression to regular cigarette smoking among nonsmoking adolescents: interactions with gender and ethnicity. *Prev Med* 2003; **36**: 455–63.
41. Sartor C. E., Xian H., Scherrer J. F., Lynskey M. T., Duncan A. E., Haber J. R. *et al.* Psychiatric and familial predictors of transition times between smoking stages: results from an offspring-of-twins study. *Addict Behav* 2008; **33**: 235–51.
42. Heron J., Barker E. D., Joinson C., Lewis G., Hickman M., Munafo M. *et al.* Childhood conduct disorder trajectories, prior risk factors and cannabis use at age 16: birth cohort study. *Addiction* 2013; **108**: 2129–38.
43. Sartor C. E., Lynskey M. T., Heath A. C., Jacob T., True W. The role of childhood risk factors in initiation of alcohol use and progression to alcohol dependence. *Addiction* 2007; **102**: 216–25.
44. Sartor C. E., Kranzler H. R., Gelernter J. Rate of progression from first use to dependence on cocaine or opioids: a cross-substance examination of associated demographic, psychiatric, and childhood risk factors. *Addict Behav* 2014; **39**: 473–9.
45. Gillespie N. A., Neale M. C., Kendler K. S. Pathways to cannabis abuse: a multi-stage model from cannabis availability, cannabis initiation and progression to abuse. *Addiction* 2009; **104**: 430–8.
46. Neale M. C., Cardon L. R. *Methodology for Genetic Studies of Twins and Families*. Dordrecht, the Netherlands: Kluwer Academic Publishers; 1992, pp. 189–97.
47. Lynskey M. T., Heath A. C., Nelson E. C., Bucholz K. K., Madden P. A. F., Slutske W. S. *et al.* Genetic and environmental contributions to cannabis dependence in a national young adult twin sample. *Psychol Med* 2002; **32**: 195–207.

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ADOLESCENT SUBSTANCE ABUSE (TA CHUNG, SECTION EDITOR)

Genetic and Environmental Interplay in Adolescent Substance Use Disorders

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Abstract Adolescent substance use is of considerable public health importance. This narrative review provides a brief background to genetically informative research methodologies and highlights key recent literature examining the interplay between genetic and environmental influences in the etiology of substance use. Twin studies have quantified the magnitude of genetic and environmental influences, and more recently, correlative and Children of Twin designs have shown environments can moderate heritability. Studies have identified a number of specific gene variants (e.g. OPRM1, DRD4, 5HTTLPR) that interact with parenting and peer influence, and the effectiveness of interventions may vary by genotype. However, little research has taken into account the stage-sequential nature of substance use. This may obscure important differences in the genetic and environmental influences, and their interplay, at the stages of escalation to problem use. Future research needs to build on existing methodologies to disentangle the complexities of progression in adolescent substance use.

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Introduction

Substance use is an area of considerable public and research importance due to the associated health and social consequences [1]. Use during adolescence is an area of particular research focus, not just because adolescence is the typical period for substance use initiation [2] but also because of the acute risks and harms [3–5] from substance use and the strong association between onset, extent of use during adolescence and risks for the subsequent development of substance dependence and related harms [6–12]. There has been a long history of research into environmental risk factors for the initiation of substance use [13] and more recently a recognition of strong genetic influences on substance use and substance use disorders, derived principally from the findings of twin studies [12, 14–18]. Advances in genotyping technologies have permitted the expansion of research on these genetic influences to the identification of specific genetic variants associated with drug use. Despite this, relatively few genetic variants have been identified as being robustly associated with substance dependence, with some notable exceptions [19–21].

The limited number of genetic variants with strong, robust associations with substance use has led to increasing focus on the interplay between genetic predispositions and environmental exposures in the etiology of substance use and escalation to problems [22–26]. Gene by environmental interplay encompasses both gene by environment interaction and gene by environment correlation. Three main categories of gene by environmental correlation have been identified. These are active correlation, whereby individuals select, modify or construct experiences that are correlated with their genetic

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predisposition; passive correlation, whereby individuals are passively exposed to environments that are correlated with their genetic predispositions; and evocative correlation, whereby the individual's genotype elicits a certain response from the environment around them [27]. Gene by Environment Interactions (G×E) occur when environmental effects on individuals vary by genotype or when the environment alters the effect a gene has on an individual's physiology [27, 28].

In this review, we focus on the recent literature exploring genetic and environmental influences on the development of substance use and dependence during adolescence. By highlighting methodological approaches, preliminary findings and the necessity of considering the stage-sequential nature of substance use, we identify areas that show the greatest promise for disentangling this complex etiology.

Genetically Informative Research and the Risk Factors for Adolescent Substance Use

The magnitude of genetic and environmental influences on substance use disorders has been broadly quantified by twin studies. Agrawal and Lynskey [17] noted that estimates of the heritability of substance dependence ranged from 0.30 to 0.70 and were broadly equivalent across individual substances. Twin methodologies allow the quantification of the magnitude of both shared environmental influences that increase twin similarity on a trait and non-shared environmental influences that do not increase similarity on a trait [27]. Shared genetic influences are typically less prominent than non-shared environmental influences in the etiology of substance dependence; although in adolescence, the shared environment may be relatively more prominent. In addition to quantifying the magnitude of genetic and shared environmental influences on substance dependence, twin studies have also demonstrated that there are likely to be genetic influences that form a vulnerability to substance use across drug classes [29, 30], although some evidence has been found for separate, albeit highly correlated, licit and illicit drug genetic factors [31].

Utilising Traditional Twin Studies to Explore Gene by Environment Interplay

Twin studies have not only allowed quantification of the magnitude of genetic effects but have also provided strong evidence that the magnitude of genetic influences is altered by the environment. For example, Boardman et al. have demonstrated the impact of public health policies, finding that the magnitude of genetic influences on smoking desistance increased following the introduction of restrictive legislation on smoking behaviours [32] and that genetic influences on daily smoking in adolescents were lower in states with more restrictive access to tobacco products [33]. Social factors also play an important role. For example, the heritability of smoking is higher in schools where the most popular students smoke [34]; genetic influences on adolescent smoking are more prominent in the presence of low parental monitoring [35]; and heritable influences on adolescent substance use are more prominent in environments characterised by high levels of peer substance use [36].

The Utility of Large, Linked Administrative Data Sets

In addition to results from twin studies, Kendler and his colleagues have recently used a novel research strategy to examine genetic influences, based on the analysis of whole population administrative data sets from Sweden, with sample sizes exceeding one million individuals. Combining official records of treatment seeking, official police contact and related information at a whole population level with information on outcomes in relatives (e.g. full siblings, cousins), they have confirmed substantial heritability for drug abuse in both males (55 %) and females (73 %), with environmental factors shared by siblings operating only in males [37]. The convergence of these findings with those from traditional twin studies, which have typically studied less severe phenotypes based on self-report symptom data, is impressive. Using a similar approach to examine concordance for drug abuse in pairs of related individuals, the authors demonstrated the importance of family environmental influences on drug abuse in sibling pairs, where those whose older sibling had recorded drug abuse had a 1.42 (95 % CI, 1.31–1.54) increased likelihood of drug abuse compared to those whose younger siblings had recorded drug abuse [38••]. Similar findings were reported for cousin pairs, who were found to be significantly more similar in recorded drug abuse if they were close in age and location when growing up [39].

In addition to confirming the importance of both (latent) genetic and environmental influences on risks for drug abuse, these authors have presented a number of analyses examining the extent to which specific, measured environmental exposures are associated with risks for drug abuse while controlling for heritable influences on such risk. For example, Giordano et al. [40], using a co-relative comparison within sibling and cousin pairs discordant for exposure to trauma, demonstrated that experiencing a second-hand traumatic event (e.g. having a parent or sibling be assaulted or die) before age 15 was associated with twice the risk for the development of drug abuse. They used a similar approach to highlight the importance of neighbourhood social deprivation on the development of drug abuse [41]. Kendler et al. [42] highlighted risks for drug abuse associated with exposure to both peer deviance and parental divorce while also implicating interactions between genetic liability for drug abuse and peer deviance and between parental divorce and peer deviance in the etiology of officially recorded drug abuse. For those with low genetic risk (determined by the

drug abuse records of relatives), living in an area with high peer deviance was associated with an increase in 28.1 cases of drug abuse per 10,000 person-years. In comparison, being at very high genetic risk in a high risk area was associated with an increase of 78.0 cases per 10,000 person-years.

The Use of Children of Twins Design

One potential liability of the traditional twin method is that any G×E will be confounded with, and therefore inflate, estimates of heritability unless measured environmental exposures are explicitly modelled [43]. One design which can model both main effects of genes and environment and gene by environmental interactions is the children of twins (CoT) design. This design allows a comparison of outcomes in (1) children at high genetic risk and high environmental risk (e.g. parent is alcohol dependent), (2) high genetic risk but reduced environmental risk (parent is not alcohol dependent but their MZ co-twin is), (3) intermediate genetic risk but reduced environmental risk (parent is not alcohol dependent but their DZ co-twin is) and (4) children at low genetic and low environmental risk (control families; both parent and co-twin are not alcohol dependent). Thus, it allows detection not only of genetic transmission but also environmental consequences of parental alcohol dependence that may depend upon offspring genotype (Gene × Shared Environment interaction [43]) or be masked by genetic nonadditivity.

Applying the CoT design to study the links between parental drug dependence (alcohol or cannabis) and onset of substance use in offspring, Waldron et al. [44••] reported that both children of parents who were drug dependent (high genetic/high environmental risk) and those of non-drug-dependent parents whose MZ co-twin was drug dependent (high genetic/low environmental risk) had elevated rates of early onset tobacco (7.30 and 2.22 times higher risk, respectively), alcohol (1.43 and 2.82 times higher risk, respectively) and cannabis use (only significant for those with high genetic/low environmental risk, where it was 3.03 times higher). While this pattern of results strongly implicates heritable influences in transmission of risks associated with parental drug dependence, further analyses also highlighted that, independent of these influences, exposure to parental divorce significantly elevated risks of early onset substance use.

Interactions of Specific Genetic Variants with Environmental Exposures

While the research above has focussed on latent genetic influences, research that has explored specific genes in relation to the environment has also found encouraging results for G×E. There are a number of considerations when selecting specific gene variants to be tested against specific environmental exposures. What is considered a large sample for environmental research may be underpowered for genetic studies, with extremely high numbers of participants required to identify strong G×E between genotypes and environmental exposures that commonly occur [45, 46], resulting in the employment of prioritisation strategies for investigation of G×E that focus on genetic variants that are common in the population; that are associated with the disorder being studied; or that are associated with individual response to the environmental factor under consideration [28, 45]. Consequently, the genetic variants that have been studied to date are those commonly studied in the mental health G×E literature.

There is some evidence that the influence of specific gene variants may be moderated by environmental exposures such as neighbourhood peer substance use, parental supervision, and parent-child attachment. For example, Daw et al. [47] studied the influence of 5HTTLPR in combination with measures of peer smoking. There was not a statistically significant main effect of neighbourhood peer cigarette use, but those with more copies of the 5HTTLPR short allele had a significantly higher hazard of smoking initiation in environments where a higher proportion of neighbourhood peers smoked. Specifically, the hazard ratio for the interaction between peer use and number of 5HTTLPR short alleles on any smoking was 3.532 ($P=0.002$) and 5.686 ($P<0.001$) for regular smoking. In addition, COMT has been found to interact with parental supervision to affect frequency of drinking at age 19 and with parental involvement to affect amount consumed at age 19 [48]. The interaction between OPRM1 and parental rule setting was also found to significantly differentiate heavy drinkers from light drinkers [49]. Further, the effect of an ADH1B variant has been found to be moderated by the drinking behaviour of peers, with a protective effect on reaching drinking milestones observed when no friends were reported to be drinking that was significantly reduced when most or all best friends were reported to be drinking [50]. Finally, higher numbers of DRD4 alleles significantly interacted with parent-child attachment to increase risk of problematic smoking (anxious attachment) and cannabis use (avoidant and anxious attachments) [51].

Genetic Variants and Response to Substance Use Interventions

A new and emerging research area is the examination of how specific genetic variants may alter response to treatment or prevention interventions. Understanding why certain interventions are successful for some people and less so for others would allow for the development of tailored intervention programmes and represent an example of potential G×E interactions. However, only recently has research begun to consider whether an individual's genetic risk can moderate the effects of participation in an intervention for substance use.

Brody et al. [52••] reported the results of two longitudinal family-centred randomised controlled trials designed to reduce alcohol use in 900 11–17 year olds. They examined whether adolescents assigned to the intervention condition who carried a dopaminergic or GABAergic susceptibility gene would demonstrate greater decreases in alcohol use than adolescents who did not carry either susceptibility gene. They found that participants within the intervention condition carrying two or three susceptibility genes evinced smaller increases in alcohol use than participants with none or only one susceptibility gene. Regression estimates for the interaction between gene and intervention (significant after adjustment for multiple comparisons) were −0.86 (SE 0.22) for GABRG1, −0 .67 (SE 0.21) for GABRA2 and −1.19 (SE 0.29) for DRD2. These findings provide support for the differential susceptibility theory [53] which suggests that the genetic make-up of some individuals may increase malleability or susceptibility to both negative and positive environmental influences. Thus, some individuals may be adversely affected by negative exposures and may also be more likely to benefit from positive environments. Therefore, inter-individual variability can exist in response to positive experiences such as programmes designed to address substance use.

Methodological Challenges of Specific Genetic Variants

Research is providing promising results for G×E and has highlighted genes that warrant further exploration. However, the selection of specific genetic variants for study has limitations. With regard to the environmental exposures selected for study, there are an extensive number of potential exposures that can be combined with the plethora of gene candidates. Sher and colleagues [54] have highlighted that the unavailability of information on the number of possible environmental influences means that there is no "environome" to compare with the genome. Distal factors that are thought to have a long-term effect (such as childhood family environment) can be differentiated from proximal exposures (such as availability), and studies of G×E in adolescent substance use may benefit from systematically classifying environment in this way. There is also a need to identify whether G×E findings remain stable at different time points. Developmental stage has previously been shown to affect gene associations [55], and specifically for adolescent substance use, it is important to confirm whether associations differ depending on the type of drug, the stage of substance use, the developmental stage of the tested population and across subgroups. Finally, despite years of candidate gene studies, there are still few replicated associations [46]. Additional lines of G×E research may identify more robust associations.

Polygenic Risk Scores

Focussing research on combinations of genes or gene systems may be a more efficient strategy for understanding G×E in substance use. The advent of genome-wide association studies (GWAS) has implicated multiple genes from multiple biological pathways in the etiology of substance use [19, 56]. However, as for other complex traits, the effect sizes of individual gene variants associated with substance use are small and fall short of accounting for the expected amount of variability based on heritability estimates [57]. Taking this into account, some researchers have moved from examining single genes for G×E to considering polygenic risk scores [58•, 59, 60]. In principle, these scores work in the same way as other risk prediction models, aggregating the effects of multiple genetic variants to produce a quantitative estimate of (genetic) risk for a particular trait for each individual.

While risk scores do have the potential to be useful and informative, careful consideration must be given to (i) methods used to develop these scores, (ii) the strength of association between the polygenic score and the outcome(s) of interest and (iii) whether the results are applicable beyond the study sample [61]. An illustrative paper on this topic used a longitudinal community sample to investigate the interaction between a polygenic risk score and age in relation to tobacco consumption [62•]. The authors demonstrated that, even when multiple genetic factors were combined, the polygenic score only accounted for a small proportion of the variance in a trait for nicotine (in their example, from 0.1 to 1 % of the variance in cigarette consumption between the ages of 14 and 24) and did not show the same relationship for alcohol. This does not rule out the use of polygenic risk scores but does serve to underline the need for realistic expectations about their likely predictive power and the importance of considering their potential contribution in combination with other, nongenetic, risk factors [63].

Considering the Stage-Sequential Nature of Drug Dependence

There are three key features of the relationship between genetic and environmental influences on the development of substance use and dependence that require further elaboration. Firstly, it is important to note that the development of substance dependence is a stage-sequential process in which a number of different transitions must first occur (see Fig. 1). Many individuals will reach different stages without progressing to dependence, but traditionally, most studies of dependence have conflated these stages. For example, the common comparison of individuals who are substance dependent against those who are not dependent often fails to distinguish between non-cases who may have never used a drug and

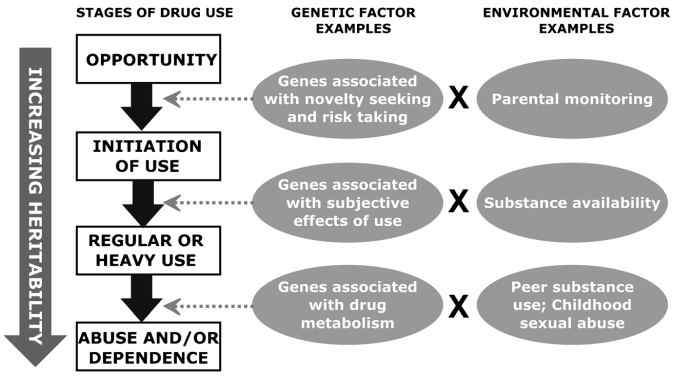

Fig. 1 Diagram of stage-sequential drug use, depicting plausible stage-specific genetic and environmental influences, and the increasing magnitude of genetic influences on later stages

those who used the drug (potentially regularly) but did not progress to dependence. There is also value in exploring variation amongst those who reach different stages, differentiating not only those who do or do not initiate use but also those who initiate use earlier or later [64]. Despite research highlighting the necessity of considering early substance use stages, this has rarely been considered, making it unclear at which stages in the development of drug dependence specific genetic or environmental influences are most prominent [65].

Secondly, research suggests that the relative strength of genetic and environmental influences on the development of substance dependence varies by both stage of substance use and by developmental age [12, 66–70]. For example, there is evidence that, in their aggregate, genetic influences are relatively weaker in younger aged samples [71] and at earlier stages (e.g. initiation) in the development of substance dependence [16] (see Fig. 1). Nonetheless, there is also evidence of substantial, albeit incomplete, overlap in the genetic influences on initiation and problem use [72–74].

A third feature that deserves further exploration is the extent to which the importance of measured genetic and environmental influences on substance dependence may vary across stages of substance involvement (see Fig. 1). Early stages of substance use, such as initiation, might be genetically influenced through personality traits such as novelty seeking [75], while genetic factors associated with sensitivity to drug effects may influence progression of use. At subsequent stages, such as drug dependence and withdrawal, genetic influences on drug metabolism may have stronger associations. Similarly, there may be distal and proximal environmental risk factors that are unique to specific stages of substance involvement, while others may act across multiple stages or show correlation while not being identical [72]. Research is also emerging into how external influences can lead to changes in gene function at a cellular level [76], and these epigenetic mechanisms have recently begun to attract interest in the field of substance use [77–79]. Speculatively, early stages of use may be influenced by pre-existing epigenetic modification (resulting, for example, from childhood stressors [80–83]) with later stages influenced by epigenetic modifications brought about by substance use. Consequently, it is expected that the use of the stage-sequential approach will reveal that observed genetic and environmental associations will alter throughout the sequence towards dependence.

Recent research utilising the stage-sequential approach has demonstrated differences in association by stage of use for both environmental and genetic factors. For example, Sartor et al. [68] reported a number of associations with environmental factors that were unique to onset of alcohol use (e.g. male gender, attention deficit hyperactivity disorder, parental divorce and maternal alcohol dependence), while others were unique to the transition from alcohol use to dependence (e.g. nicotine dependence, cannabis abuse, generalised anxiety disorder). Similar results have been obtained for genetic risk factors. Belsky et al. [58•] reported that a multi-locus genetic risk score, derived from the results of meta-analyses for nicotine dependence, was unrelated to initiation of tobacco use but was significantly associated with increased risks for daily tobacco use, more rapid progression from initiation to heavy use, increased risks for the development of nicotine dependence and reduced likelihood of successful cessation. The literature on substance use behaviour trajectories has typically studied behaviour change that occurs between substance use stages, and modelling development and changes in substance use over time has found that GABRA2 is associated with an increase in drunkenness between ages 18–19, suggested to be due to the enhanced independence related to reaching adulthood [84]. Similarly, OPRM1 has been found to differentiate those who were light drinkers from those who had progressed to moderate drinking in participants followed up over 6 years (participants on average aged 14.3 at start of study) [49].

In summary, studies utilising the stage-sequential approach are likely to reveal differential genetic influences at different stages, with substance-specific genes more likely to operate at later stages [85]. Different environmental influences are also likely to operate at each stage, and research into trajectories suggests that environmental change should be incorporated into analysis [84]. Taking this approach to research is key for gaining a comprehensive understanding of G×E in the development of adolescent substance use and to reaching meaningful conclusions about opportunities for intervention.

Conclusions

The research currently available highlights the role of genetic and environmental influences and, importantly, their interplay in the development of adolescent substance use and provides intriguing avenues for potential interventions. The evidence shows promising leads with regard to gene variants that warrant further exploration, the composite effects of genes on substance use and the importance of the type and timing of environmental influences in shaping adolescent substance use behaviour.

Genetic and environmental interplay continues to be a promising avenue for exploration in order to understand the underlying causes of adolescent substance use and progression and future research will need to apply the optimal strategies for investigating this area [28, 45, 61], demonstrate the replicability of findings and overcome issues around the measurement of phenotypes. Incorporation of new methodologies provides the best opportunity to overcome the barriers to these aims. As discussed in this review, the innovative use of noninvasive methods for research through administrative data sets [38••, 39, 41, 42] shows promise for analysis of large samples with measured phenotypes at relatively low cost and effort. Combining such methods with a stage-sequential approach to understanding genetic and environmental influences on drug involvement and response to intervention may be key to disentangling the complex etiology of one of the great public health issues of our time.

Compliance with Ethics Guidelines

Conflict of Interest Lindsey A. Hines, Katherine I. Morley, Clare Mackie and Michael Lynskey declare that they have no conflict of interest.

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References

Papers of particular interest, published recently, have been highlighted as:
- • Of importance
- •• Of major importance

1. Degenhardt L, Whiteford H, Hall WD. The global burden of disease projects: what have we learned about illicit drug use and dependence and their contribution to the global burden of disease? Drug Alcohol Rev. 2014;33(1):4–12.
2. Kandel DB, Logan JA. Patterns of drug use from adolescence to young adulthood: I. Periods of risk for initiation, continued use, and discontinuation. Am J Public Health. 1984;74(7):660–6.
3. Movig KL et al. Psychoactive substance use and the risk of motor vehicle accidents. Accid Anal Prev. 2004;36(4):631–6.
4. Tapert SF et al. Adolescent substance use and sexual risk-taking behavior. J Adolesc Health. 2001;28(3):181–9.
5. Hingson RW, Zha W, Weitzman ER. Magnitude of and trends in alcohol-related mortality and morbidity among U.S. college students ages 18–24, 1998–2005. J Stud Alcohol Drugs Suppl. 2009;2009(16):12–20.
6. Hingson RW et al. Age of drinking onset and injuries, motor vehicle crashes, and physical fights after drinking and when not drinking. Alcohol Clin Exp Res. 2009;33(5):783–90.
7. Lynskey MT et al. Early-onset alcohol-use behaviors and subsequent alcohol-related driving risks in young women: a twin study. J Stud Alcohol Drugs. 2007;68(6):798–804.
8. Grant JD et al. Associations of alcohol, nicotine, cannabis and drug use/dependence with educational attainment: evidence from cotwin-control analyses. Alcohol Clin Exp Res. 2012;36(8):1412–20.
9. Hingson RW, Heeren T, Winter MR. Age at drinking onset and alcohol dependence: age at onset, duration, and severity. Arch Pediatr Adolesc Med. 2006;160(7):739–46.
10. Chen CY, Storr CL, Anthony JC. Early-onset drug use and risk for drug dependence problems. Addict Behav. 2009;34(3):319–22.
11. Behrendt S et al. Transitions from first substance use to substance use disorders in adolescence: is early onset associated with a rapid escalation? Drug Alcohol Depend. 2009;99(1–3):68–78.
12. Lynskey MT et al. An Australian twin study of cannabis and other illicit drug use and misuse, and other psychopathology. Twin Res Hum Genet. 2012;15(5):631–41.
13. Hawkins JD, Catalano RF, Miller JY. Risk and protective factors for alcohol and other drug problems in adolescence and early adulthood: implications for substance abuse prevention. Psychol Bull. 1992;112(1):64–105.
14. Lynskey MT et al. Genetic and environmental contributions to cannabis dependence in a national young adult twin sample. Psychol Med. 2002;32(2):195–207.
15. Verweij KJ et al. Genetic and environmental influences on cannabis use initiation and problematic use: a meta-analysis of twin studies. Addiction. 2010;105(3):417–30.
16. Lynskey MT, Agrawal A, Heath AC. Genetically informative research on adolescent substance use: methods, findings, and challenges. J Am Acad Child Adolesc Psychiatry. 2010;49(12):1202–14.
17. Agrawal A, Lynskey MT. Are there genetic influences on addiction: evidence from family, adoption and twin studies. Addiction. 2008;103(7):1069–81.
18. Kendler KS. What psychiatric genetics has taught us about the nature of psychiatric illness and what is left to learn. Mol Psychiatry. 2013;18(10):1058–66.
19. Agrawal A et al. The genetics of addiction—a translational perspective. Transcult Psychiatry. 2012;2:e140.

20. Cao J, Hudziak JJ, Li D. Multi-cultural association of the serotonin transporter gene (SLC6A4) with substance use disorder. Neuropsychopharmacology. 2013;38(9):1737–47.
21. Lubke G et al. The CHRNA5/A3/B4 gene cluster and tobacco, alcohol, cannabis, inhalants and other substance use initiation: replication and new findings using mixture analyses. Behav Genet. 2012;42(4):636–46.
22. Young-Wolff KC, Enoch M-A, Prescott CA. The influence of gene–environment interactions on alcohol consumption and alcohol use disorders: a comprehensive review. Clin Psychol Rev. 2011;31(5):800–16.
23. Dick DM, Kendler KS. The impact of gene-environment interaction on alcohol use disorders. Alcohol Res Curr Rev. 2012;34(3):318–24.
24. Moffitt TE, Caspi A, Rutter M. Measured gene-environment interactions in psychopathology: concepts, research strategies, and implications for research, intervention, and public understanding of genetics. Perspect Psychol Sci. 2006;1(1):5–27.
25. Rutter M, Moffitt TE, Caspi A. Gene-environment interplay and psychopathology: multiple varieties but real effects. J Child Psychol Psychiatry. 2006;47(3–4):226–61.
26. Meyers JL, Dick DM. Genetic and environmental risk factors for adolescent-onset substance use disorders. Child Adolesc Psychiatr Clin N Am. 2010;19(3):465–77.
27. Plomin R, DeFries JC, Knopik VS, Neiderhiser JM. Behavioral genetics. 6th ed. New York: Worth Publishers; 2013.
28. Moffitt TE, Caspi A, Rutter M. Strategy for investigating interactions between measured genes and measured environments. Arch Gen Psychiatry. 2005;62(5):473–81.
29. Tsuang MT et al. Co-occurrence of abuse of different drugs in men: the role of drug-specific and shared vulnerabilities. Arch Gen Psychiatry. 1998;55(11):967–72.
30. Kendler KS et al. Specificity of genetic and environmental risk factors for use and abuse/dependence of cannabis, cocaine, hallucinogens, sedatives, stimulants, and opiates in male twins. Am J Psychiatry. 2003;160(4):687–95.
31. Kendler KS, Myers J, Prescott CA. Specificity of genetic and environmental risk factors for symptoms of cannabis, cocaine, alcohol, caffeine, and nicotine dependence. Arch Gen Psychiatry. 2007;64(11):1313–20.
32. Boardman JD et al. Population composition, public policy, and the genetics of smoking. Demography. 2011;48(4):1517–33.
33. Boardman JD. State-level moderation of genetic tendencies to smoke. Am J Public Health. 2009;99(3):480–6.
34. Boardman JD et al. Do schools moderate the genetic determinants of smoking? Behav Genet. 2008;38(3):234–46.
35. Dick DM et al. Parental monitoring moderates the importance of genetic and environmental influences on adolescent smoking. J Abnorm Psychol. 2007;116(1):213–8.
36. Agrawal A et al. Peer substance involvement modifies genetic influences on regular substance involvement in young women. Addiction. 2010;105(10):1844–53.
37. Kendler KS et al. Genetic and family and community environmental effects on drug abuse in adolescence: a Swedish national twin and sibling study. Am J Psychiatry. 2014;171(2):209–17.
38.•• Kendler KS et al. Within-family environmental transmission of drug abuse: a Swedish national study. JAMA Psychiatry. 2013;70(2):235–42. **Taking a novel approach to the study of genetic effects, this study provides an unprecedently large sample for analysis. What is striking is that this methodology overcomes a number of the limitations of traditional heritability studies (albeit bringing new study limitations), but findings support those of more traditional designs.**
39. Kendler KS et al. Environmental influences on familial resemblance for drug abuse in first-cousin pairs: a Swedish national study. Psychol Med. 2014;44(2):371–9.
40. Giordano GN et al. Unexpected adverse childhood experiences and subsequent drug use disorder: a Swedish population study (1995–2011). Addiction. 2014;109(7):1119–27.
41. Kendler KS et al. The causal nature of the association between neighborhood deprivation and drug abuse: a prospective national Swedish co-relative control study. Psychol Med. 2014;44(12):2537–46.
42. Kendler KS et al. Peer deviance, parental divorce, and genetic risk in the prediction of drug abuse in a nationwide Swedish sample: evidence of environment-environment and gene-environment interaction. JAMA Psychiatry. 2014;71(4):439–45.
43. Heath AC et al. Gene-environment interaction effects on behavioral variation and risk of complex disorders: the example of alcoholism and other psychiatric disorders. Twin Res. 2002;5(1):30–7.
44.•• Waldron M et al. Parental separation and early substance involvement: results from children of alcoholic and cannabis dependent twins. Drug Alcohol Depend. 2014;134:78–84. **This study utilizes the Children of Twins design, which avoids some limitations of the classical twin design and allows consideration of potential gene by environment interaction. This paper demonstrates the importance of parental separation as a risk factor over and above the genetic and environmental effects of parental drug dependence.**
45. Uher R. Gene-environment interactions in common mental disorders: an update and strategy for a genome-wide search. Soc Psychiatry Psychiatr Epidemiol. 2014;49(1):3–14.
46. Duncan LE, Keller MC. A critical review of the first 10 years of candidate gene-by-environment interaction research in psychiatry. Am J Psychiatry. 2011;168(10):1041–9.
47. Daw J et al. The interactive effect of neighborhood peer cigarette use and 5HTTLPR genotype on individual cigarette use. Addict Behav. 2014;39(12):1804–10.
48. Laucht M et al. Catechol-O-methyltransferase Val158 Met genotype, parenting practices and adolescent alcohol use: testing the differential susceptibility hypothesis. J Child Psychol Psychiatry. 2012;53(4):351–9.
49. van der Zwaluw CS et al. Different trajectories of adolescent alcohol use: testing gene-environment interactions. Alcohol Clin Exp Res. 2014;38(3):704–12.
50. Olfson E et al. An ADH1B variant and peer drinking in progression to adolescent drinking milestones: evidence of a gene-by-environment interaction. Alcohol Clin Exp Res. 2014;38(10):2541–9.
51. Olsson CA et al. Gene-environment interaction in problematic substance use: interaction between DRD4 and insecure attachments. Addict Biol. 2013;18(4):717–26.
52.•• Brody GH, Chen YF, Beach SR. Differential susceptibility to prevention: GABAergic, dopaminergic, and multilocus effects. J Child Psychol Psychiatry. 2013;54(8):863–71. **The differential susceptibility hypothesis proposes that, as well as genetic influences priming individuals for greater reactivity to negative environmental exposures, such as exposure to trauma, these genetic influences may also confer greater susceptibility to positive environmental exposures, including treatment and other interventions. This paper is one of the first to study this in the context of interventions for adolescent substance use.**
53. Belsky J, Pluess M. Beyond diathesis stress: differential susceptibility to environmental influences. Psychol Bull. 2009;135(6):885–908.
54. Sher KJ et al. Consilient research approaches in studying gene × environment interactions in alcohol research. Addict Biol. 2010;15(2):200–16.
55. Dick DM et al. How phenotype and developmental stage affect the genes we find: GABRA2 and impulsivity. Twin Res Hum Genet. 2013;16(3):661–9.

56. Kapoor M et al. Genome-wide survival analysis of age at onset of alcohol dependence in extended high-risk COGA families. Drug Alcohol Depend. 2014;142:56–62.
57. Manolio TA et al. Finding the missing heritability of complex diseases. Nature. 2009;461(7265):747–53.
58.• Belsky DW et al. Polygenic risk and the developmental progression to heavy, persistent smoking and nicotine dependence: evidence from a 4-decade longitudinal study. JAMA Psychiatry. 2013;70(5):534–42. **This study is one of the few to address the stage-sequential nature of drug use while incorporating a polygenic risk score. Results suggested that genetic risk was unrelated to smoking initiation, but was related to conversion to regular smoking, speed of transition to regular smoking, and cessation failure.**
59. McGeary JE et al. Predictors of relapse in a bupropion trial for smoking cessation in recently-abstinent alcoholics: preliminary results using an aggregate genetic risk score. Subst Abus. 2012;6:107–14.
60. Salvatore JE et al. Polygenic scores predict alcohol problems in an independent sample and show moderation by the environment. Genes (Basel). 2014;5(2):330–46.
61. Wray NR et al. Pitfalls of predicting complex traits from SNPs. Nat Rev Genet. 2013;14(7):507–15.
62.• Vrieze SI, McGue M, Iacono WG. The interplay of genes and adolescent development in substance use disorders: leveraging findings from GWAS meta-analyses to test developmental hypotheses about nicotine consumption. Hum Genet. 2012;131(6):791–801. **This paper tests whether SNPs are specific to smoking or are relevant for more pervasive behavioural systems. Previous research has found no association between genes associated with smoking initiation and genes associated with cigarettes smoked per day, and this paper finds that nicotine-specific genetic risk becomes more important in adulthood than in adolescence. Effect sizes are small, but the paper highlights important concepts.**
63. Hall WD, Mathews R, Morley KI. Being more realistic about the public health impact of genomic medicine. PLoS Med. 2010;7(10):e1000347. doi:10.1371/journal.pmed.1000347
64. Heath AC et al. Estimating two-stage models for genetic influences on alcohol, tobacco or drug use initiation and dependence vulnerability in twin and family data. Twin Res. 2002;5(2):113–24.
65. Nelson EC et al. ANKK1, TTC12, and NCAM1 polymorphisms and heroin dependence: importance of considering drug exposure. JAMA Psychiatry. 2013;70(3):325–33.
66. Kendler KS et al. A population-based twin study in women of smoking initiation and nicotine dependence. Psychol Med. 1999;29(2):299–308.
67. Sartor CE et al. Genetic and environmental influences on the rate of progression to alcohol dependence in young women. Alcohol Clin Exp Res. 2008;32(4):632–8.
68. Sartor CE et al. The role of childhood risk factors in initiation of alcohol use and progression to alcohol dependence. Addiction. 2007;102(2):216–25.
69. Distel MA et al. Age moderates non-genetic influences on the initiation of cannabis use: a twin-sibling study in Dutch adolescents and young adults. Addiction. 2011;106(9):1658–66.
70. Heath AC et al. Genetic and social determinants of initiation and age at onset of smoking in Australian twins. Behav Genet. 1999;29(6):395–407.
71. Kendler KS et al. Recent advances in the genetic epidemiology and molecular genetics of substance use disorders. Nat Neurosci. 2012;15(2):181–9.
72. Kendler KS et al. Genetic and environmental risk factors in the aetiology of illicit drug initiation and subsequent misuse in women. Br J Psychiatry. 1999;175:351–6.
73. Neale MC et al. Extensions to the modeling of initiation and progression: applications to substance use and abuse. Behav Genet. 2006;36(4):507–24.
74. Gillespie NA, Neale MC, Kendler KS. Pathways to cannabis abuse: a multi-stage model from cannabis availability, cannabis initiation and progression to abuse. Addiction. 2009;104(3):430–8.
75. Laucht M et al. Novelty seeking involved in mediating the association between the dopamine D4 receptor gene exon III polymorphism and heavy drinking in male adolescents: results from a high-risk community sample. Biol Psychiatry. 2007;61(1):87–92.
76. Dupont C, Armant DR, Brenner CA. Epigenetics: definition, mechanisms and clinical perspective. Semin Reprod Med. 2009;27(5):351–7.
77. Wong CC, Mill J, Fernandes C. Drugs and addiction: an introduction to epigenetics. Addiction. 2011;106(3):480–9.
78. van der Knaap LJ et al. Catechol-O-methyltransferase gene methylation and substance use in adolescents: the TRAILS study. Genes Brain Behav. 2014;13(7):618–25.
79. Nielsen DA et al. Epigenetics of drug abuse: predisposition or response. Pharmacogenomics. 2012;13(10):1149–60.
80. Essex MJ et al. Epigenetic vestiges of early developmental adversity: childhood stress exposure and DNA methylation in adolescence. Child Dev. 2013;84(1):58–75.
81. Dube SR et al. Childhood abuse, neglect, and household dysfunction and the risk of illicit drug use: the adverse childhood experiences study. Pediatrics. 2003;111(3):564–72.
82. Anda RF et al. Adverse childhood experiences and smoking during adolescence and adulthood. JAMA. 1999;282(17):1652–8.
83. Dube SR et al. Adverse childhood experiences and the association with ever using alcohol and initiating alcohol use during adolescence. J Adolesc Health. 2006;38(4):444.e1–e10.
84. Dick DM et al. Genetic influences on alcohol use across stages of development: GABRA2 and longitudinal trajectories of drunkenness from adolescence to young adulthood. Addict Biol. 2014;19(6):1055–64.
85. Sartor CE et al. Common genetic contributions to alcohol and cannabis use and dependence symptomatology. Alcohol Clin Exp Res. 2010;34(3a):545–54.

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Full length article

Onset of opportunity to use cannabis and progression from opportunity to dependence: Are influences consistent across transitions?

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ABSTRACT

Background: There is a developing body of research looking at cannabis use opportunity, but little research examining timing of opportunity to use cannabis.
Aims: Identify factors associated with (1) earlier opportunity to use cannabis and (2) faster progression from opportunity to cannabis dependence.
Method: Cross-sectional study of 3824 Australian twins and siblings, measuring age of onset of cannabis use opportunity and DSM-IV cannabis dependence. Survival analysis identified factors associated with faster progression to opportunity or dependence.
Results: Factors associated with both speed of progression to opportunity and dependence were conduct disorder (opportunity HR 5.57, 95%CI 1.52–20.47; dependence HR 2.49, 95%CI 1.91–3.25), parental drug problems (opportunity HR 7.29, 95%CI 1.74–30.62; dependence HR 3.30, 95%CI 1.63–6.69), weekly tobacco use (opportunity HR 8.57, 95%CI 3.93–18.68; dependence HR 2.76, 95% CI 2.10–3.64), and female gender (opportunity HR 0.69, 95%CI 0.64–0.75; dependence HR 0.44, 95%CI 0.34–0.55). Frequent childhood religious attendance (HR 0.74, 95%CI 0.68–0.80), parental conflict (HR 1.09, 95%CI 1.00–1.18), parental alcohol problems (HR 1.19, 95%CI 1.08–1.30) and childhood sexual abuse (HR 1.17, 95%CI 1.01–1.34) were uniquely associated with transition to opportunity. Depressive episode (HR 1.44, 95%CI 1.12–1.85), tobacco dependence (HR 1.36, 95%CI 1.04–1.78), alcohol dependence (HR 2.64, 95%CI 1.53–4.58), other drug use (HR 2.10, 95%CI 1.64–2.69) and other drug dependence (HR 2.75, 95%CI 1.70–4.43) were uniquely associated with progression to dependence.
Conclusion: The profile of factors associated with opportunity to use cannabis and dependence only partially overlaps, suggesting targeting of interventions may benefit from being tailored to the stages of drug use.

1. Introduction

Cannabis is widely used, with cumulative lifetime incidence of use estimated to range from 6%-20% in Europe, 3–11% in the Middle East and Africa, and to exceed 40% in the US and New Zealand (Degenhardt et al., 2008). Lifetime prevalence of cannabis use in Australian adolescents has been estimated at 60% (Patton et al., 2002). Although many individuals use cannabis infrequently and without experiencing problems, globally an estimated 13.1 million individuals meet criteria for cannabis dependence, contributing 10.3% of the illicit drug use global burden of disease (Degenhardt et al., 2014). It is estimated 10–16% of cannabis users develop dependence (Anthony, 2006), but before progressing to dependence individuals must pass through a number of preceding stages. Examining the multiple stages of drug use before dependence develops is necessary for gaining a comprehensive understanding of factors involved in drug use, and for identifying opportunities for early intervention (Hines et al., 2015a).

The first stage of drug involvement is having the opportunity to use (regardless of whether the individual uses the drug or not),

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which forms the "exposure opportunity" in the epidemiology of drug use (Wagner and Anthony, 2002). Opportunity is required for use to occur, and forms an individual's earliest necessary condition from which they are at risk of developing cannabis dependence. Recent research indicates the majority of adolescents who have an opportunity to use cannabis progress to initiation of use (Caris et al., 2009; Lopez-Quintero and Neumark, 2015; Pinchevsky et al., 2011), making the opportunity to use an important target for intervention (Neumark et al., 2012).

There is a developing body of research looking at the opportunity to use. Factors associated with opportunity to use cannabis include using alcohol, using tobacco and the combination of alcohol and tobacco use (Caris et al., 2009; Neumark et al., 2012; Wagner and Anthony, 2002). In Chile and the US, males have been found to be slightly more likely than females to have a chance to use cannabis (Caris et al., 2009; Van Etten and Anthony, 1999), but these gender differences have not been consistently observed (Wells et al., 2011). Childhood religious practices are associated with decreased likelihood of cannabis use opportunity (Chen et al., 2004), and those with externalising behaviour problems have been found to be more likely to have a cannabis use opportunity (Neumark et al., 2012; Reboussin et al., 2015). Perhaps unsurprisingly given that first cannabis use opportunity typically occurs in late childhood or early adolescence, lower parental involvement and higher levels of coercive discipline have been found to be associated with increased likelihood of cannabis use opportunity (Chen et al., 2005). The effect of parenting continues throughout adolescence, with those who reported low parental monitoring in high school more likely to have cannabis use opportunity once they started college (Pinchevsky et al., 2011).

Amongst this existing evidence, there is little research examining the timing of opportunity to use cannabis. The study of transitions, and the timing of these transitions, can provide unique insights into influences on substance use (Behrendt et al., 2012; Hines et al., 2015b; Sartor et al., 2009, 2008), but only a limited number of factors have been studied in relation to speed of transition to cannabis use opportunity (with earlier opportunity representing a faster transition) These have focussed on early childhood behaviours, with disruptive behaviour early in school in males and better reading scores in females associated with earlier cannabis use opportunity (Storr et al., 2011). Similarly, no research to date has explored whether there is overlap between factors associated with earlier opportunity and those associated with the speed of progression to dependence. These include other substance use (Behrendt et al., 2009), some mental health factors (Behrendt et al., 2011) and gender (Ridenour et al., 2006; Wittchen et al., 2008). Exploring speed of transition to cannabis opportunity will determine whether risk factors for dependence are already exerting influence on drug use behaviours at the start of an individual's cannabis involvement, which has utility for improving understanding of how dependence develops (Hines et al., 2015a). Applying survival analysis methodology to this area allows for quantification of time to cannabis use opportunity and from opportunity to dependence, and identification of what factors may impact upon the speed of these transitions.

This paper aims to:

1. Identify factors associated with earlier opportunity to use cannabis.
2. Identify factors associated with progression from cannabis use opportunity to cannabis dependence.
3. Determine whether factors associated with opportunity to use cannabis are also associated with more rapid progression from first opportunity to dependence.

2. Methods

2.1. Sample

The sample was drawn from the Australian Twin Registry. From a pool of twin pairs born between 1972 and 1979, 3348 MZ and DZ twins and 476 of their siblings (mean age at time of interview = 32.1, SD 3.04, range 21–46) completed the interview component of a study of cannabis and other drug misuse. A full description of the study methodology and of the characteristics of participants has been published previously (Lynskey et al., 2012).

2.2. Assessment

Participants were assessed through computer-assisted telephone interviews which collected information on socio-demographics, childhood experiences, drug use and common mental health disorders, including conduct disorder and major depressive disorder, assessed using the SSAGA-OZ interview (Bucholz et al., 1994; Heath et al., 1997). The SSAGA-OZ is a validated measure of mental health using DSM-IV criteria, and includes assessment of cannabis and other drug abuse and dependence. Specific measures used in the current analyses are described below.

2.3. Measures

2.3.1. Outcome measures.

2.3.1.1. Opportunity to use cannabis. Participants were asked "have you ever been offered, or had the opportunity to use cannabis, even if you did not use it at the time? How old were you the first time?" Of 3,824 individuals interviewed, 3,798 provided information on whether or not they had ever had the opportunity to use cannabis. Of these, 85% ($N = 3399$) reported they had an opportunity to use cannabis. A continuous measure of age of first opportunity was used for both survival analysis models.

2.3.1.2. Cannabis dependence. Participants were classified as meeting lifetime criteria for DSM-IV cannabis dependence (American Psychiatric Association, 2000) if they reported three or more of the following symptoms occurring within the same 12 month period: using cannabis a greater number of times/greater amount than was intended, tolerance, wanting to cut down/stop use, spending so much time obtaining/using/recovering from the effects of cannabis the participant had little time for anything else, reducing important activities as a result of cannabis use, continuing use despite it worsening health/emotional problems. Withdrawal was not included as it was not part of DSM-IV criteria for cannabis dependence. Participants were also asked the age at which they first experienced three or more of these symptoms occurring within a 12 month period.

Of those reporting lifetime opportunity to use cannabis, 10.9% ($N = 371$) met criteria for cannabis dependence, and a continuous measure of age at onset of cannabis dependence was used in survival analysis.

2.3.2. Covariates.

2.3.2.1. Gender. Gender was determined through self-report.

2.3.2.2. Parental alcohol problems. Parental alcohol problems were determined through participant self-report of their mother or father experiencing problems with health/family/job/police/other as a result of drinking, or their mother or father drinking excessively.

2.3.2.3. Parental drug problems. Parental drug problems were determined through participant self-report of their mother or father experiencing problems with health/family/job/police/other as a result of drug use, or the participant reporting they felt their mother or father had a problem with drugs.

2.3.2.4. Parental conflict. Parental conflict was determined by participant responses to the questions "how often did your parents fight or argue in front of you?" and "how much conflict and tension was there between your parents?" Both questions focused on the period when the participant was aged 6–13. Participants reporting parents 'sometimes' or 'always' fought or argued, or 'a lot' or 'some' conflict/tension, were coded as experiencing high parental conflict.

2.3.2.5. Single parent family. Single parent family was determined by participants' report of whether their mother or father was absent. Interviewers recorded whether participants lived with their mother/mother figure and/or their father/father figure for at least 4 full years between 6 and 13.

2.3.2.6. Strict parenting. Strict parenting was determined through participants response to the items "In your opinion, when you were 6–13, was your mother/mother figure more strict than most mothers?" and " In your opinion, when you were 6–13, was your father/father figure more strict than most fathers?". Those who endorsed either of these items were classified as having experienced strict parenting.

2.3.2.7. Childhood sexual abuse. Childhood sexual abuse was recorded for individuals who reported being forced into sexual intercourse or any other forms of sexual activity before age 18. Self-reported age of sexual abuse onset was used to create a time varying covariate for sexual abuse.

2.3.2.8. Frequent childhood religious attendance. Frequent childhood religious attendance was determined through participant self-report of their frequency of attendance at religious services between ages 6 and 13. Participants were coded as frequently attending religious services if they reported attendance more than once a week, once a week, once or twice a month or every couple of months.

2.3.2.9. Conduct disorder. Conduct disorder was determined by participant self-report of at least 3 of the 15 DSM-IV criteria (American Psychiatric Association, 2000) occurring within the same 12-month period, prior to age 18. Participants' self-reported age of onset of 3 symptoms occurring within a 12 month period was used to create a time varying covariate for conduct disorder.

2.3.2.10. Depressive episode. Depressive episode was recorded if participants reported a two week period where they were more irritable than usual (if under age 18 at the time), felt depressed/down/sad/blue/discouraged, or had a lot less interest in things. Self-reported age of the first occurring depressive episode was used to make time varying covariates for survival analysis.

2.3.2.11. Weekly tobacco use. Weekly tobacco use was measured through the interview item "Was there ever a time in your life when you smoked cigarettes at least once a week for at least two months in a row?" Self-reported age of onset of weekly tobacco use was used to make time varying covariates for survival analysis.

2.3.2.12. Tobacco dependence. Tobacco dependence was measured through participants reporting 3 or more of the DSM-IV tobacco dependence criteria (American Psychiatric Association, 2000) occurring within a 12 month period. Self-reported age of onset of tobacco dependence was used to make time varying covariates for survival analysis.

2.3.2.13. Monthly alcohol use. Monthly alcohol use was measured through the interview item "At what age did you start to drink regularly—that is, drinking at least once a month for 6 months or more?" Self-reported age of onset of monthly alcohol use was used to make time varying covariates for survival analysis.

2.3.2.14. Alcohol dependence. Alcohol dependence was measured through participants reporting 3 or more of the DSM-IV alcohol dependence criteria (American Psychiatric Association, 2000) occurring within a 12 month period. Self-reported age of onset of alcohol dependence was used to make time varying covariates for survival analysis.

2.3.4.15. Other drug use. Other drug use was recorded if participants reported lifetime non-prescribed use of any of the following: cocaine (all forms), stimulants, opiates and major painkillers, sedatives, hallucinogens, dissociatives, solvents or inhalants. Self-reported age of drug use onset was used to create a time varying covariate for first other drug use.

2.3.2.16. Other drug dependence. Other drug dependence was recorded if participants reported lifetime dependence on any of the following: cocaine (all forms), stimulants, opiates and major painkillers, sedatives, hallucinogens, dissociatives, solvents and inhalants. Participants were classified as meeting lifetime criteria for DSM-IV drug dependence if they reported 3 or more of the 7 DSM-IV symptoms of dependence (American Psychiatric Association, 2000) occurring within the same 12 month period. Self-reported age of onset of dependence was used to create a time varying covariate for other drug dependence. This covariate was only included in the model of progression to the development of dependence.

2.4. Statistical analyses

All analyses were conducted in Stata statistical software version 11 (StataCorp, 2009). Two separate Cox proportional hazard models were fitted to the data to test the association between a number of potential associated factors and speed of progression from (1) birth to opportunity to use cannabis and (2) opportunity to use cannabis to the development of cannabis dependence. Both were assessed as time in years. Details of the two Cox Proportional Hazards models are provided below:

Model one: To identify factors associated with hazard of the opportunity to use cannabis survival data (time in years, starting from birth) were used for analysis of 3,798 participants who had provided information on opportunity to use cannabis. Failure event was opportunity to use cannabis, and 3398 failure events were observed (one participant was excluded from analysis, see description below). Due to missing covariate data, 3,763 participants were included in the final model (3367 failure events).

Model two: To identify factors associated with hazard of the development of dependence following the opportunity to use cannabis survival data (time in years, starting from age of first opportunity to use cannabis) were used for analysis of 2,593 participants who had reported their age of opportunity to use cannabis and who had also reported lifetime cannabis use (those who had not reported lifetime cannabis use were removed from the model in order to avoid the inverse association that would exist between never-use of cannabis and cannabis dependence; additionally, one participant was omitted as their recorded age of dependence was earlier than recorded age of opportunity). The failure event was

cannabis dependence, and 371 failures were observed. Due to missing covariate data, 2,565 participants were included in the final model (363 failure events).

Person year data sets were constructed providing a separate row of participant data for each year from birth for model 1, and for each year from age of opportunity for model 2. In order to account for multiple participants experiencing failures events in the same year, the Efron adjustment for survival ties (Efron, 1977) was applied. Participants were right-censored at age of interview.

Factors described above were included in the model. Time varying measures were produced for conduct disorder, monthly alcohol use, alcohol dependence, weekly tobacco use, tobacco dependence, other drug use, other drug dependence, childhood sexual abuse, and depressive episode. These variables were coded as present for each year after the age of onset, and were only included in the model if they were positive prior to the onset of cannabis use opportunity for model one, or prior to the onset of dependence for model two (e.g., if age at opportunity to use cannabis was 13, then conduct disorder with an age of onset of 14 was coded as absent prior to the onset of opportunity).

To minimise the likelihood that the effect of childhood covariates where the specified time periods were ages 6–13 (parental conflict, single parent family, strict parenting, frequent childhood religious attendance) may have occurred after the point of cannabis use opportunity, any individuals who reported use opportunity before the age of 6 were removed from model one. This resulted in the observations of only one participant being removed from the model. Huber-White analysis for clustered data was implemented to adjust for the non-independence of observations from members of a twin pair. The assumption of proportional hazards was assessed through tests of Schoenfeld residuals and modelling of the interaction of covariates with time in the analysis (represented as '_t') ($P = \leq 0.05$). Any variables found to violate the proportional hazards assumption were reparameterized via modelling interactions between the variable and time in the analysis, resulting in an extended Cox Proportional Hazards model.

Analyses on the transition from opportunity to first use of cannabis could not be conducted due to insufficient variation in this measure (the majority of participants progressed from opportunity to first use 0 or 1 years after having the opportunity to use, data available on request).

3. Results

3.1. Sample, survival data and the proportional hazards assumption

Comparisons between those who did and did not report lifetime cannabis use opportunity, and those who did and did not progress to cannabis use following opportunity, show these groups differ on the majority of the covariates tested within the survival models (see Tables 1). Mean age of first cannabis use opportunity was 17.6 (s.d. 3.2) and the mean age of cannabis dependence 21.4 (s.d. 4.1). The mean survival time for the participants in the cannabis use opportunity model was 19.1 years (s.d. 5.1) (age at opportunity, or for those who did not report opportunity, age at interview.) This figure is higher than the mean opportunity age as individuals who have not experienced opportunity by the point of interview are also included in the survival model, with their age at time of interview in place of age of opportunity. The mean survival time for participants in the cannabis dependence model was 13.4 years (s.d. 4.9) (time from opportunity to dependence, or for those who did not develop dependence, time from opportunity to age at interview). This figure is higher than may be expected from the mean dependence age as individuals who have not developed dependence by the point of interview are also included in the survival time, with their time from opportunity to age at interview in place of time to dependence.

All covariates were tested for breach of the proportional hazards assumption, as outlined in the methods section. The following did not satisfy the proportional hazards assumption for the opportunity to use model and therefore the interaction term between the factor and analysis time was modelled in the cannabis use opportunity analysis (Bellera et al., 2010): conduct disorder, parental drug problems, weekly tobacco use and monthly alcohol use. Similarly, for the cannabis dependence analysis the following variables had the interaction with analysis time modelled in the analysis: parental drug problems, alcohol dependence and other drug use.

3.2. Factors uniquely associated with opportunity to use cannabis

Results from the Cox proportional hazards model for transition to opportunity to use cannabis are presented in Table 3. Conduct disorder, high parental conflict, parental alcohol problems, parental drug problems, childhood sexual abuse and weekly tobacco use were associated with increased hazard of earlier opportunity to use cannabis. Frequent childhood religious attendance and female gender were independently associated with slower transition to cannabis use opportunity.

3.3. Factors uniquely associated with progression to cannabis dependence

Results from the Cox proportional hazards model for transition from opportunity to use cannabis to dependence are presented in Table 3. Conduct disorder, parental drug problems, weekly tobacco use, depressive episode, tobacco dependence, alcohol dependence, other drug use and other drug dependence were associated with increased hazard of faster transition cannabis dependence. Female gender was independently associated with slower transition to cannabis dependence.

3.4. Factors consistently associated across transitions

Factors associated with increased hazard of both earlier cannabis use opportunity and faster progression to cannabis dependence were conduct disorder, parental drug problems, and weekly tobacco use (see Table 3). Female gender was associated with slower progression to both cannabis use opportunity and dependence.

4. Discussion

This paper identifies a number of factors uniquely associated with the transition to cannabis use opportunity and with the transition from opportunity to cannabis dependence, and several factors that increase hazards of both these transitions. Parental conflict, parental alcohol problems and childhood sexual abuse were uniquely associated with faster transition to opportunity, whilst frequent childhood religious attendance was associated with slower transition to opportunity. Depressive episode, tobacco dependence, alcohol dependence, other drug use and other drug dependence were uniquely associated with faster progression from opportunity to dependence. Conduct disorder, parental drug problems and weekly tobacco use were associated with faster progression to both opportunity and from opportunity and dependence, with female gender associated with slower transition for both.

Exploring a broad range of factors has identified similarities and inconsistencies with the existing literature. Frequent childhood religious attendance, associated with reduced likelihood of cannabis use opportunity, was consistent with existing literature

Table 1
Comparison of characteristics of those who reported no lifetime cannabis use opportunity with those who reported lifetime cannabis use opportunity, and those who reported cannabis use opportunity and did not progress to use with those who did progress to use (proportions and odds ratios).

	No opportunity to use cannabis *N* = 399 N (%)	Opportunity to use cannabis *N* = 3399 N (%)	Odds Ratio (95% CI)	Opportunity but did not initiate cannabis use *N* = 805 N (%)	Opportunity and initiated cannabis use *N* = 2593 N (%)	Odds ratio (95% CI)
Female gender	326 (81.7)	2099 (61.8)	0.36 (0.28–0.47)	535 (66.5)	1563 (60.3)	0.77 (0.65–0.91)
Conduct disorder	4 (1.0)	320 (9.4)	10.30 (3.82–27.76)	24 (2.98)	296 (11.4)	4.21 (2.76–6.43)
Depressive episode	185 (46.5)	1636 (48.3)	1.08 (0.87–1.32)	374 (46.5)	1262 (48.9)	1.10 (0.94–1.29)
High parental conflict[†]	128 (32.1)	1272 (37.4)	1.27 (1.02–1.58)	257 (31.9)	1015 (39.2)	1.37 (1.16–1.62)
Parental alcohol problems	57 (14.3)	895 (26.3)	2.15 (1.61–2.87)	183 (22.7)	712 (27.5)	1.29 (1.07–1.55)
Parental drug problems	5 (1.3)	125 (3.7)	3.03 (1.23–7.46)	19 (2.36)	106 (4.1)	1.78 (1.09–2.92)
Single parent family[†]	14 (3.5)	203 (6.0)	1.75 (1.01–3.03)	48 (6.0)	155 (6.0)	1.00 (0.72–1.40)
Strict parenting[†]	183 (45.9)	1672 (49.2)	1.14 (0.93–1.41)	371 (46.1)	1301 (50.3)	1.18 (1.01–1.38)
Frequent childhood religious attendance[†]	299 (74.9)	1981 (58.3)	0.47 (0.37–0.59)	512 (63.6)	1468 (56.6)	0.75 (0.63–0.88)
Childhood sexual abuse	20 (5.1)	303 (9.0)	1.86 (1.17–2.95)	48 (6.0)	255 (9.9)	1.73 (1.26–2.38)
Weekly tobacco use	30 (7.5)	1493 (44.0)	9.65 (6.61–14.09)	110 (13.7)	1382 (53.4)	7.23 (5.83–8.96)
Tobacco dependence	15 (3.8)	946 (27.8)	9.89 (5.87–16.65)	50 (6.2)	895 (34.5)	7.97 (5.92–10.73)
Monthly alcohol use	274 (68.7)	3182 (93.6)	6.72 (5.22–8.65)	682 (84.7)	2500 (96.5)	4.90 (3.69–6.51)
Alcohol dependence	19 (4.8)	928 (27.3)	7.51 (4.71–11.98)	85 (10.6)	843 (32.5)	4.08 (3.21–5.18)
Other drug use	49 (12.3)	1623 (47.8)	6.54 (4.81–8.88)	140 (17.4)	1483 (57.2)	6.36 (5.21–7.75)
Other drug dependence	0 (0.0)	178 (5.2)	–	5 (0.6)	173 (6.7)	11.51 (4.71–28.10)

[†] When participant was aged 6–13 years old.

Table 2
Mean age (standard deviation) of behaviour onsets of those who reported no lifetime cannabis use opportunity with those who reported lifetime cannabis use opportunity, and those who reported cannabis use opportunity and did not progress to use with those who did progress to use.

	No opportunity to use cannabis *N* = 399	Opportunity to use cannabis *N* = 3399	Opportunity but did not initiate cannabis use *N* = 805	Opportunity and initiated cannabis use *N* = 2593
Conduct disorder	12.5 (s.d. 4.20)	14.2 (s.d. 2.31)	14.0 (s.d. 2.88)	14.2 (s.d. 2.26)
Depressive episode	22.4 (s.d. 6.26)	21.8 (s.d. 6.42).	21.8 (s.d. 6.51)	21.8 (s.d. 6.40)
Childhood sexual abuse	11.9 (s.d. 4.56)	11.1 (s.d. 4.68)	10.2 (s.d. 4.58)	11.3 (s.d. 4.69)
Weekly tobacco use	17.2 (s.d. 2.64)	17.3 (s.d. 3.44).	18.3 (s.d. 3.58)	17.3 (s.d. 3.42)
Tobacco dependence	23.8 (s.d. 7.77)	21.9 (s.d. 4.47).	23.5 (s.d. 4.62)	21.8 (s.d. 4.45)
Monthly alcohol use	20.4 (s.d. 3.72)	18.0 (s.d. 2.57).	19.1 (s.d. 3.14)	17.7 (s.d. 2.31)
Alcohol dependence	22.6 (s.d. 4.79)	22.5 (s.d. 4.20).	22.7 (s.d. 4.27)	22.5 (s.d. 4.19)
Other drug use	23.7 (s.d. 6.20)	21.6 (s.d. 4.26)	21.9 (s.d. 5.45)	21.6 (s.d. 4.13)
Other drug dependence	0 (0.0)	23.0 (s.d. 4.52)	25.8 (s.d. 3.90)	22.9 (s.d. 4.52)

Table 3
Hazard ratios (95%CI) from cox regression models: factor associated with earlier opportunity to use cannabis, and for progression from opportunity to use cannabis to cannabis dependence.

Covariate	Transition to cannabis use opportunity *N* = 3763		Transition to cannabis dependence *N* = 3367	
	Unadjusted HR (95% CI)	Adjusted HR (95% CI)	Unadjusted HR (95% CI)	Adjusted HR (95% CI)
Female gender	0.70*** (0.65–0.75)	0.69*** (0.64–0.75)	0.50*** (0.40–0.62)	0.44*** (0.34–0.55)
Conduct disorder[a]	[b]7.54*** (2.39–23.76)	[b]5.57** (1.52–20.47)	4.57*** (3.63–5.75)	2.49*** (1.91–3.25)
Depressive episode[a]	1.04 (0.93–1.17)	0.98 (0.87–1.10)	1.95*** (1.55–2.42)	1.44*** (1.12–1.85)
High parental conflict[†]	1.09* (1.01–1.18)	1.09* (1.00–1.18)	1.16 (0.94–1.44)	1.02 (0.79–1.31)
Parental alcohol problems	1.27*** (1.16–1.38)	1.19*** (1.08–1.30)	1.29** (1.03–1.62)	1.11 (0.86–1.43)
Parental drug problems	[b]8.26** (2.12–32.15)	[b]7.29** (1.74–30.62)	[b]4.14*** (2.07–8.27)	[b]3.30*** (1.63–6.69)
Single parent family[†]	1.30** (1.10–1.53)	1.13 (0.95–1.35)	1.60* (1.11–2.32)	1.19 (0.78–1.81)
Strict parenting[†]	1.03 (0.96–1.10)	1.02 (0.95–1.09)	1.32** (1.07–1.62)	1.11 (0.88–1.39)
Frequent childhood religious attendance[†]	0.72*** (0.66–0.78)	0.74*** (0.68–0.80)	0.86 (0.69–1.07)	0.84 (0.67–1.06)
Childhood sexual abuse[a]	1.25** (1.08–1.42)	1.17* (1.01–1.34)	1.98*** (1.49–2.64)	1.35 (0.95–1.92)
Weekly tobacco use[a]	[b]10.17*** (5.00–20.71)	[b]8.57*** (3.93–18.68)	3.98*** (3.12–5.07)	2.76*** (2.10–3.64)
Tobacco dependence[a]	1.82* (1.29–2.56)	0.89 (0.63–1.25)	2.77*** (2.18–3.52)	1.36* (1.04–1.78)
Monthly alcohol use[a]	[b]1.65 (0.78–3.50)	[b]0.75 (0.34–1.64)	1.03 (0.75–1.41)	0.94 (0.69–1.30)
Alcohol dependence[a]	1.79*** (1.29–2.48)	1.26 (0.89–1.78)	[b]2.94*** (1.69–5.12)	[b]2.64*** (1.53–4.58)
Other drug use[a]	1.31** (1.05–1.65)	1.20 (0.94–1.52)	2.76*** (2.22–3.42)	2.10*** (1.64–2.69)
Other drug dependence[a]	–	–	5.70*** (4.77–10.94)	2.75*** (1.70–4.43)

Note: Cannabis dependence *N* = 363 (due to missing covariate data). HR = Hazard ratio.
*$P < 0.05$ **$P < 0.01$ ***$P < 0.001$.
[a] Time Varying Covariate.
[b] interaction with _t included in the model to account for breach of the proportional hazards assumption.
[†] When participant was aged 6 – 13 years old.

(Chen et al., 2004). In contrast to prior literature (Miller et al., 2000) this protective effect did not extend to dependence. Depressive episode was associated with increased speed of transition to dependence, which is consistent with emerging findings of an asso-

ciation between depression and cannabis use disorders (Feingold et al., 2015; Pacek et al., 2013), but was not found to be associated with earlier opportunity to use cannabis. This may be due to the age of depressive episode onset occurring after age of cannabis use opportunity for the majority of participants. Previous research has reported that childhood adversity and sexual abuse are associated with other drug use opportunity and cannabis dependence (Benjet et al., 2013; Duncan et al., 2008) but, while the present analyses identified an association between childhood sexual abuse and earlier cannabis use opportunity, there was no association between childhood sexual abuse and progression from opportunity to dependence. Differences between the present findings and existing research may be due to the relatively novel exploration of speed of transitions between stages rather than the likelihood of outcomes, which has been the focus of much existing research.

The identification of tobacco, alcohol and other drug involvement as factors associated with progression from opportunity to dependence suggests that a pattern of poly-use emerges. Although alcohol use has previously been found to be associated with early onset of cannabis use (Coffey et al., 2000) it was not associated with opportunity to use cannabis in the present analyses, which may partially reflect the high prevalence of monthly alcohol use in the current sample. The comparatively rarer outcomes of tobacco dependence, other drug use and other drug dependence were found to be associated with increased speed of progression to cannabis dependence. The use of both tobacco and cannabis has been frequently observed (Agrawal et al., 2012, 2010; Hindocha et al., 2015), and regular cigarette smokers are more likely to report earlier cannabis use opportunity (Agrawal et al., 2013). Present results strongly supported this finding, and extend it to show weekly tobacco use and dependence were significantly associated with speed of progression to cannabis dependence. The observed association between cannabis dependence and tobacco may be due to a number of factors including shared genetic and environmental influences, the co-administration of tobacco and cannabis, and smoking habituation (Agrawal et al., 2012).

A number of factors were associated with both transitions studied. Female gender was associated with slower progression to both opportunity and dependence. It is interesting to note that gender differences held across both transitions given that previous research has found males more likely to have opportunity to use cannabis, but has found these gender differences do not extend to the transition into drug use once opportunity has occurred (van Etten et al., 1999). Similarly, weekly tobacco use was associated with increased hazard of both cannabis use opportunity and progression to cannabis dependence, consistent with existing findings relating to dependence (Wagner and Anthony, 2002). Conduct disorder was associated with faster progression to both opportunity and dependence, echoing previous research showing disruptive or aggressive behaviour in both males and females is associated with earlier opportunity to use cannabis (Storr et al., 2011). Parental drug problems were significantly associated with a more rapid transition to both opportunity and dependence, in line with existing research relating to opportunity (Benjet et al., 2013). This factor most clearly demonstrated changes in the magnitude of effect size between transitions, and given the especially strong association with opportunity to use cannabis it is plausible that parental drug problems facilitate an environment in which drug access is increased, whether this is indirectly or directly through parents. Alternatively, cannabis availability has previously been shown to be influenced by genetic effects (Gillespie et al., 2009), and the present finding may represent a genetic liability to creating drug use opportunities.

The pattern of results presented in this paper demonstrates that the influence of factors differs throughout the stages of drug use progression. Research relating to early onset of drug use often calls for earlier detection and intervention (Chen et al., 2009), and the current findings have two key implications for prevention. Firstly, as factors play different roles across drug involvement, interventions may benefit from tailoring to stages of drug use. Secondly, targeting of interventions may improve by considering the consistency and differences in associated factors across the stages of drug use. Using the results of the present study may facilitate identification of populations who will benefit from targeted or indicated prevention strategies (National Research Council (US) and Institute of Medicine (US) Committee on the Prevention of Mental Disorders and Substance Abuse Among Children, Youth, and Young Adults: Research Advances and Promising Interventions, 2009).

There are certain considerations required in interpretation of this work. Firstly, analyses were conducted on retrospective self-report data, introducing the possibility of recall bias. This is a viable method of data collection (Darke, 1998; Sartor et al., 2011), and indeed recall of early experience with cannabis has been found to be especially reliable (Johnson and Mott, 2001), but as the analyses rely on accurate recall of age of onset of a number of behaviours the work would benefit from replication in longitudinal cohorts.

Secondly, analyses of the progression from opportunity to cannabis use initiation were not possible, as timing of transitions was only available as time in years, and there was not enough variation in the speed of this transition to allow for analysis (the majority of participants progressed to use within 1 year after having the opportunity to use, data available on request). Thirdly, selected covariates measured occurrence within an age range (6–13), and consequently may have occurred prior to the age of opportunity to use cannabis for a small number of individuals. Fourthly, while the prevalence of lifetime cannabis use in this sample was relatively high at 68.2% (Lynskey et al., 2012) it is important to note this estimate is consistent with previous estimates from the Australian young adult population (Australian Institute of Health and Welfare, 2014). Finally, interpretation of these analyses should be in light of the twin and sibling sample used, as there is some residual uncertainty about whether inferences from twin data have external validity with respect to what might be found in general population samples (Vitaro et al., 2009). Analyses were adjusted for clustering effects using the Huber-White estimator, which was selected over other potential analyses that can be conducted to explore within twin/sibling frailties as the most parsimonious method.

Consideration of multiple stages of drug use from non-use to dependence allows identification of factors uniquely associated with specific transitions. The current results demonstrate that different factors are influential at different stages of the development of cannabis dependence. Additionally, the differences and consistencies in factors across the stages of drug use provide an insight into which similarities and differences we may expect to see occurring through the transitions towards dependence. The findings have implications for substance use prevention efforts, as both the targeting of interventions as well as the interventions themselves may benefit from being tailored to stages of drug use.

Conflict of interest

AA has previously received peer-reviewed funding from ABMRF/Foundation for Alcohol Research which receives partial support from the brewing industry.

JS is a researcher and clinician and has worked with a range of types of treatment and rehabilitation service-providers. He has also worked with pharmaceutical companies to seek to identify new or improved treatments, and also with a range of governmental and non-governmental organisations. His employer (King's College London) is registering intellectual property on an innovative medication development with which JS is involved (not relevant

to cannabis), and JS has been named in a patent registration by a Pharma company as inventor of a potential novel overdose resuscitation product (not relevant to cannabis). A fuller account of JS's interests is on his personal web-page of the Addictions Department at http://www.kcl.ac.uk/ioppn/depts/addictions/people/hod.aspx . JS is also supported by the National Institute for Health Research (NIHR) Biomedical Research Centre for Mental Health at South London and Maudsley NHS Foundation Trust and King's College London.

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Author contributions

Lindsey A. Hines developed hypotheses, ran analysis, writing of paper.

Katherine I. Morley supervised analyses, comments throughout drafting.

John Strang comments once paper was drafted.

Arpana Agrawal comments once paper was drafted, input to design of analyses.

Elliot C. Nelson comments once paper was drafted.

Dixie Statham: organiser of original twin study, comments once paper was drafted.

Nicholas G. Martin: organiser of original twin study, comments once paper was drafted.

Michael T. Lynskey: substantive input at planning stages, comments throughout drafting.

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References

Agrawal, A., Budney, A.J., Lynskey, M.T., 2012. The co-occurring use and misuse of cannabis and tobacco: a review. Addiction 107, 1221–1233, http://dx.doi.org/10.1111/j.1360-0443.2012.03837.x.

Agrawal, A., Madden, P.A.F., Martin, N.G., Lynskey, M.T., 2013. Do early experiences with cannabis vary in cigarette smokers?? Drug Alcohol Depend. 128, 255–259, http://dx.doi.org/10.1016/j.drugalcdep.2012.09.002.

Agrawal, A., Silberg, J.L., Lynskey, M.T., Maes, H.H., Eaves, L.J., 2010. Mechanisms underlying the lifetime co-occurrence of tobacco and cannabis use in adolescent and young adult twins. Drug Alcohol Depend. 108, 49–55, http://dx.doi.org/10.1016/j.drugalcdep.2009.11.016.

American Psychiatric Association, 2000. Diagnostic And Statistical Manual Of Mental Disorders: DSM-IV-TR, Text Revision, 4th Ed. American Psychiatric Association, Washington, DC.

Anthony, J.C., 2006. The epidemiology of cannabis dependence. In: Roffman, R.A., Stephens, R.S. (Eds.), Cannabis Dependence: Its Nature, Consequences, and Treatment. Cambridge University Press, New York, pp. 58–105.

Australian Institute of Health and Welfare, 2014. National Drug Strategy Household Survey detailed report: 2013. Drug statistics series no. 28. Cat. no. PHE 183. AIHW, Canberra.

Behrendt, S., Beesdo-Baum, K., Hofler, M., Perkonigg, A., Buhringer, G., Lieb, R., Wittchen, H.U., 2012. The relevance of age at first alcohol and nicotine use for initiation of cannabis use and progression to cannabis use disorders. Drug Alcohol Depend. 123, 48–56, http://dx.doi.org/10.1016/j.drugalcdep.2011.10.013.

Behrendt, S., Beesdo-Baum, K., Zimmermann, P., Hofler, M., Perkonigg, A., Buhringer, G., Lieb, R., Wittchen, H.U., 2011. The role of mental disorders in the risk and speed of transition to alcohol use disorders among community youth. Psychol. Med. 41, 1073–1085, http://dx.doi.org/10.1017/s0033291710001418.

Behrendt, S., Wittchen, H.U., Hofler, M., Lieb, R., Beesdo, K., 2009. Transitions from first substance use to substance use disorders in adolescence: is early onset associated with a rapid escalation?? Drug Alcohol Depend. 99, 68–78, http://dx.doi.org/10.1016/j.drugalcdep.2008.06.014.

Bellera, C.A., MacGrogan, G., Debled, M., Lara, C.T., de Brouste, V., Mathoulin-Pélissier, S., 2010. Variables with time-varying effects and the Cox model: some statistical concepts illustrated with a prognostic factor study in breast cancer. BMC Med. Res. Methodol. 10, 20, http://dx.doi.org/10.1186/1471-2288-10-20.

Benjet, C., Borges, G., Medina-Mora, M.E., Méndez, E., 2013. Chronic childhood adversity and stages of substance use involvement in adolescents. Drug Alcohol Depend. 131, 85–91, http://dx.doi.org/10.1016/j.drugalcdep.2012.12.002.

Bucholz, K.K., Cadoret, R., Cloninger, C.R., Dinwiddie, S.H., Hesselbrock, V.M., Nurnberger, J., Reich, T., Schmidt, I., Schuckit, M.A., 1994. A new, semi-structured psychiatric interview for use in genetic linkage studies: a report on the reliability of the SSAGA. J. Stud. Alcohol Drugs 55, 149.

Caris, L., Wagner, F.A., Ríos-Bedoya, C.F., Anthony, J.C., 2009. Opportunities to use drugs and stages of drug involvement outside the United States : evidence from the Republic of Chile. Drug Alcohol Depend. 102, 30–34, http://dx.doi.org/10.1016/j.drugalcdep.2008.12.004.

Chen, C.-Y., Dormitzer, C.M., Bejarano, J., Anthony, J.C., 2004. Religiosity and the earliest stages of adolescent drug involvement in seven countries of Latin America. Am. J. Epidemiol. 159, 1180–1188, http://dx.doi.org/10.1093/aje/kwh151.

Chen, C.-Y., Storr, C.L., Anthony, J.C., 2009. Early-onset drug use and risk for drug dependence problems. Addict. Behav. 34, 319–322, http://dx.doi.org/10.1016/j.addbeh.2008.10.021.

Chen, C.Y., Storr, C.L., Anthony, J.C., 2005. Influences of parenting practices on the risk of having a chance to try cannabis. Pediatrics 115, 1631–1639, http://dx.doi.org/10.1542/peds.2004-1926.

Coffey, C., Lynskey, M., Wolfe, R., Patton, G.C., 2000. Initiation and progression of cannabis use in a population-based Australian adolescent longitudinal study. Addiction 95, 1679–1690, http://dx.doi.org/10.1046/j.1360-0443.2000.951116798.x.

Darke, S., 1998. Self-report among injecting drug users: a review. Drug Alcohol Depend. 51, 253–263, http://dx.doi.org/10.1016/s0376-8716(98) 00028-3.

Degenhardt, L., Chiu, W.-T., Sampson, N., Kessler, R.C., Anthony, J.C., Angermeyer, M., Bruffaerts, R., de Girolamo, G., Gureje, O., Huang, Y., Karam, A., Kostyuchenko, S., Lepine, J.P., Mora, M.E.M., Neumark, Y., Ormel, J.H., Pinto-Meza, A., Posada-Villa, J., Stein, D.J., Takeshima, T., Wells, J.E., 2008. Toward a global view of alcohol, tobacco, cannabis, and cocaine use: findings from the WHO World Mental Health Surveys. PLoS Med. 5, e141, http://dx.doi.org/10.1371/journal.pmed.0050141.

Degenhardt, L., Whiteford, H., Hall, W.D., 2014. The Global burden of disease projects: what have we learned about illicit drug use and dependence and their contribution to the global burden of disease?? Drug Alcohol Rev. 33, 4–12, http://dx.doi.org/10.1111/dar.12088.

Duncan, A.E., Sartor, C.E., Scherrer, J.F., Grant, J.D., Heath, A.C., Nelson, E.C., Jacob, T., Bucholz, K.K., 2008. The association between cannabis abuse and dependence and childhood physical and sexual abuse: evidence from an offspring of twins design. Addiction 103, 990–997, http://dx.doi.org/10.1111/j.1360-0443.2008.02210.x.

Efron, B., 1977. The efficiency of Cox's Likelihood Function for censored data. J. Am. Stat. Assoc., 557–565, http://dx.doi.org/10.2307/2286217.

Feingold, D., Weiser, M., Rehm, J., Lev-Ran, S., 2015. The association between cannabis use and mood disorders: a longitudinal study. J. Affective Disord. 172, 211–218, http://dx.doi.org/10.1016/j.jad.2014.10.006.

Gillespie, N.A., Neale, M.C., Kendler, K.S., 2009. Pathways to cannabis abuse: a multi-stage model from cannabis availability, cannabis initiation and progression to abuse. Addiction 104, 430–438, http://dx.doi.org/10.1111/j.1360-0443.2008.02456.x.

Heath, A.C., Bucholz, K.K., Madden, P.A., Dinwiddie, S.H., Slutske, W.S., Bierut, L.J., Statham, D.J., Dunne, M.P., Whitfield, J.B., Martin, N.G., 1997. Genetic and environmental contributions to alcohol dependence risk in a national twin sample: consistency of findings in women and men. Psychol. Med. 27, 1381–1396.

Hindocha, C., Shaban, N.D.C., Freeman, T.P., Das, R.K., Gale, G., Schafer, G., Falconer, C.J., Morgan, C.J.A., Curran, H.V., 2015. Associations between cigarette smoking

and cannabis dependence: a longitudinal study of young cannabis users in the United Kingdom. Drug Alcohol Depend. 148, 165–171, http://dx.doi.org/10.1016/j.drugalcdep.2015.01.004.
Hines, L.A., Morley, K.I., Mackie, C., Lynskey, M., 2015a. Genetic and environmental interplay in adolescent substance use disorders. Curr. Addict. Rep. 2, 122–129, http://dx.doi.org/10.1007/s40429-015-0049-8.
Hines, L.A., Morley, K.I., Strang, J., Agrawal, A., Nelson, E.C., Statham, D., Martin, N.G., Lynskey, M.T., 2015b. The association between speed of transition from initiation to subsequent use of cannabis and later problematic cannabis use, abuse and dependence. Addict. Abingdon Engl. 110, 1311–1320, http://dx.doi.org/10.1111/add.12963.
Johnson, T.P., Mott, J.A., 2001. The reliability of self-reported age of onset of tobacco, alcohol and illicit drug use. Addict. Abingdon Engl. 96, 1187–1198, http://dx.doi.org/10.1080/09652140120060770.
Lopez-Quintero, C., Neumark, Y., 2015. Prevalence and determinants of resistance to use drugs among adolescents who had an opportunity to use drugs. Drug Alcohol Depend. 149, 55–62, http://dx.doi.org/10.1016/j.drugalcdep.2015.01.015.
Lynskey, M.T., Agrawal, A., Henders, A., Nelson, E.C., Madden, P.A.F., Martin, N.G., 2012. An Australian twin study of cannabis and other illicit drug use and misuse, and other psychopathology. Twin Res. Hum. Genet. 15, 631–641, http://dx.doi.org/10.1017/thg.2012.41.
Miller, L., Davies, M., Greenwald, S., 2000. Religiosity and substance use and abuse among adolescents in the National Comorbidity Survey. J. Am. Acad. Child Adolesc. Psychiatry 39, 1190–1197, http://dx.doi.org/10.1097/00004583-200009000-00020.
National Research Council (US) and Institute of Medicine (US) Committee on the Prevention of Mental Disorders and Substance Abuse Among Children, Youth, and Young Adults: Research Advances and Promising Interventions, 2009. Preventing Mental, Emotional, and Behavioral Disorders Among Young People: Progress and Possibilities, The National Academies Collection: Reports funded by National Institutes of Health National Academies Press (US), Washington (DC).
Neumark, Y., Lopez-Quintero, C., Bobashev, G., 2012. Drug use opportunities as opportunities for drug use prevention: Bogota, Colombia a case in point. Drug Alcohol Depend. 122, 127–134, http://dx.doi.org/10.1016/j.drugalcdep.2011.09.022.
Pacek, L.R., Martins, S.S., Crum, R.M., 2013. The bidirectional relationships between alcohol, cannabis, co-occurring alcohol and cannabis use disorders with major depressive disorder: results from a national sample. J. Affective Disord. 148, 188–195, http://dx.doi.org/10.1016/j.jad.2012.11.059.
Patton, G.C., Coffey, C., Carlin, J.B., Degenhardt, L., Lynskey, M., Hall, W., 2002. Cannabis use and mental health in young people: cohort study. BMJ 325, 1195–1198, http://dx.doi.org/10.1136/bmj.325.7374.1195.
Pinchevsky, G.M., Arria, A.M., Caldeira, K.M., Garnier-Dykstra, L.M., Vincent, K.B., O'Grady, K.E., 2011. Marijuana exposure opportunity and initiation during college: parent and peer influences. Prev. Sci. 13, 43–54, http://dx.doi.org/10.1007/s11121-011-0243-4.
Reboussin, B.A., Ialongo, N.S., Green, K.M., 2015. Influences of behavior and academic problems at school entry on marijuana use transitions during adolescence in an African–American sample. Addict. Behav. 41, 51–57, http://dx.doi.org/10.1016/j.addbeh.2014.09.030.
Ridenour, T.A., Lanza, S.T., Donny, E.C., Clark, D.B., 2006. Different lengths of times for progressions in adolescent substance involvement. Addict. Behav. 31, 962–983, http://dx.doi.org/10.1016/j.addbeh.2006.03.015.
Sartor, C.E., Agrawal, A., Lynskey, M.T., Bucholz, K.K., Madden, P.A., Heath, A.C., 2009. Common genetic influences on the timing of first use for alcohol, cigarettes, and cannabis in young African–American women. Drug Alcohol Depend. 102, 49–55, http://dx.doi.org/10.1016/j.drugalcdep.2008.12.013.
Sartor, C.E., Bucholz, K.K., Nelson, E.C., Madden, P.A.F., Lynskey, M.T., Heath, A.C., 2011. Reporting bias in the association between age at first alcohol use and heavy episodic drinking. Alcohol. Clin. Exp. Res. 35, 1418–1425, http://dx.doi.org/10.1111/j.1530-0277.2011.01477.x.
Sartor, C.E., Xian, H., Scherrer, J.F., Lynskey, M.T., Duncan, A.E., Haber, J.R., Grant, J.D., Bucholz, K.K., Jacob, T., 2008. Psychiatric and familial predictors of transition times between smoking stages: results from an offspring-of-twins study. Addict. Behav. 33, 235–251, http://dx.doi.org/10.1016/j.addbeh.2007.09.002.
Storr, C.L., Wagner, F.A., Chen, C.Y., Anthony, J.C., 2011. Childhood predictors of first chance to use and use of cannabis by young adulthood. Drug Alcohol Depend. 117, 7–15, http://dx.doi.org/10.1016/j.drugalcdep.2010.12.023.
Van Etten, M.L., Anthony, J.C., 1999. Comparative epidemiology of initial drug opportunities and transitions to first use: marijuana, cocaine, hallucinogens and heroin. Drug Alcohol Depend. 54, 117–125.
van Etten, M.L., Neumark, Y.D., Anthony, J.C., 1999. Male-female differences in the earliest stages of drug involvement. Addiction 94, 1413–1419, http://dx.doi.org/10.1046/j.1360-0443.1999.949141312.x.
Vitaro, F., Brendgen, M., Arseneault, L., 2009. The discordant MZ-twin method: one step closer to the holy grail of causality. Int. J. Behav. Dev. 33, 376–382, http://dx.doi.org/10.1177/0165025409340805.
Wagner, F.A., Anthony, J.C., 2002. Into the world of illegal drug use: exposure opportunity and other mechanisms linking the use of alcohol, tobacco, marijuana, and cocaine. J. Epidemiol., 918–925 http://dx.doi.org/10.1093/aje/155.10.918.
Wells, J.E., Haro, J.M., Karam, E., Lee, S., Lepine, J.P., Medina-Mora, M.E., Nakane, H., Posada, J., Anthony, J.C., Cheng, H., Degenhardt, L., Angermeyer, M., Bruffaerts, R., de Girolamo, G., de Graaf, R., Glantz, M., Gureje, O., 2011. Cross-national comparisons of sex differences in opportunities to use alcohol or drugs, and the transitions to use. Subst. Use Misuse 46, 1169–1178, http://dx.doi.org/10.3109/10826084.2011.553659.
Wittchen, H.U., Behrendt, S., Hofler, M., Perkonigg, A., Lieb, R., Buhringer, G., Beesdo, K., 2008. What are the high risk periods for incident substance use and transitions to abuse and dependence? Implications for early intervention and prevention. Int. J. Methods Psychiatry Res. 17 (Suppl. 1), S16–S29, http://dx.doi.org/10.1002/mpr.254.